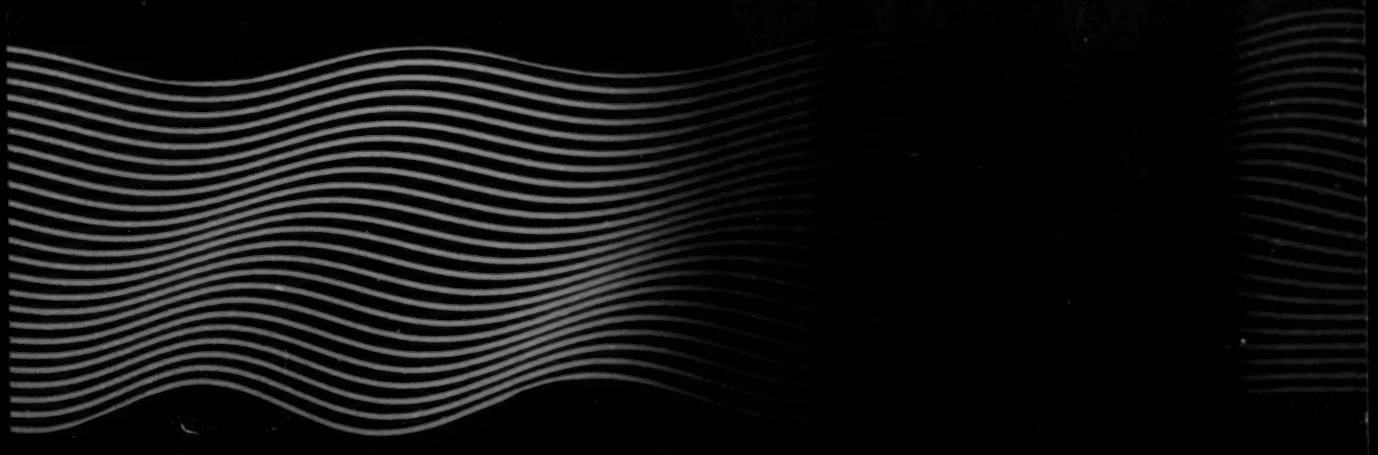
National Research Council Canada

Conseil national de recherches Canada

NRC · CNRC

Canadian Journal of Fisheries and Aquatic Sciences

Journal canadien des sciences halieutiques et aquatiques



ISSN 0706-652X / CJFSDX 54(2) 239–502 (1997)

The *Canadian Journal of Fisheries and Aquatic Sciences*, previously published as *Contributions to Canadian Biology* 1901–1925, *Contributions to Canadian Biology and Fisheries* 1926–1934, *Journal of the Biological Board of Canada* 1934–1937, and *Journal of the Fisheries Research Board of Canada* 1938–1979, is issued monthly to form annual volumes of 12 numbers plus supplements.

Editorial policy: The *Journal* publishes original research articles, critical reviews, perspectives (essays of opinion or hypothesis), and comments. Papers may concern cells, organisms, populations, ecosystems, or processes that affect aquatic production systems, and they should lead to identifiable conclusions or synthesis, which variously may amplify, modify, question, or redirect accumulated knowledge embodied in contemporary perceptions of a particular state of fisheries and aquatic sciences. They should demonstrate clearly a contribution to knowledge beyond the confirmation state. Originality should relate to more than the particular (a certain year, place, taxon, or chemical compound) such that existing understanding is reformulated or extended.

Manuscripts for publication should be submitted to **Dr. D.G. Cook**, Canadian Journal of Fisheries and Aquatic Sciences, NRC Research Press, National Research Council of Canada, Ottawa, ON K1A 0R6, Canada (613-993-2209; fax: 613-952-7656; e-mail: journal.cjfas@nrc.ca).

Submissions in either English or French are acceptable. The information must be original, that is, not copyrighted, published, or submitted elsewhere except in abstract form or unless by written consent of the Editor. NRC accepts no responsibility for statements or opinions expressed by a contributor. Acceptance of an advertisement, announcement, or other material does no imply endorsement by either the *Journal*'s Editors or NRC. The use of proprietary names does not imply endorsement of the product or company.

Correspondence concerning accepted manuscripts and corrected proofs should be sent to NRC Research Press, National Research Council of Canada, Ottawa, ON K1A 0R6, Canada.

Instructions to Authors are published once a year in the first issue of each volume. Copies are available from NRC Research Press or from our Web site. The **Publication Policy** of NRC Research Press is also available at our Web site:
http://www.nrc.ca/cisti/journals/rj.html

Microfilm: Issues from 1934 through the current volume can be purchased on 16- or 35-mm microfilm. Photocopies of individual articles or issues can be purchased from University Microfilms International, 330 North Zeeb Road, Ann Arbor, MI 48106, U.S.A.

U.S. periodical postage paid at Champlain, N.Y., and at additional mailing offices. Attention U.S. POSTMASTER: Send address changes to **Canadian Journal of Fisheries and Aquatic Sciences**, c/o USACAN Media Distribution Center, P.O. Box 2888, Plattsburg, NY 12901, U.S.A.

Publié sans interruption depuis 1901, le *Journal canadien des sciences halieutiques et aquatiques* a paru sous plusieurs titres : *Contributions to Canadian Biology* 1901–1925, *Contributions to Canadian Biology and Fisheries* 1926–1934, *Journal of the Biological Board of Canada* 1934–1937, et *Journal of the Fisheries Research Board of Canada* 1938–1979. Le *Journal* paraît tous les mois, formant ainsi un volume de 12 numéros, et offre en plus jusqu'à deux suppléments par année.

Politique de rédaction : Le journal publie des articles fondés sur une recherche originale, des critiques, des essais portant sur une opinion ou une hypothèse (perspectives) et des commentaires. Les textes peuvent avoir trait aux cellules, aux organismes, aux populations, aux écosystèmes ou aux processus qui influent sur les systèmes de production aquatique. Ils doivent aboutir à des conclusions ou synthèses précises qui, de diverse façons, peuvent accroître, modifier, remettre en question ou réorienter le bagage actuel de connaissances et perceptions dans une discipline donnée des sciences aquatiques. Ils doivent clairement démontrer qu'ils contribuent aux connaissances en faisant plus de corroborer des faits. L'originalité doit dépasser le caractère particulier (une année, un endroit, un taxon ou un composé chimique donnée) et tenir à une épuration ou une reformulation des connaissances actuelles.

Les auteurs doivent adresser leurs **manuscrits** à **D.G. Cook**, Journal canadien des sciences halieutiques et aquatiques, Presses scientifiques du CNRC, Conseil national de recherches du Canada, Ottawa, ON K1A 0R6, Canada (613-993-2209); télécopieur : 613-952-7656; courrier électronique : journal.cjfas@nrc.ca).

Les contributions peuvent être en anglais ou en français. Elles doivent être originales, c.-à.-d. qu'elles ne doivent pas avoir fait l'objet d'un copyright, avoir été publiées ou soumises ailleurs, sauf sous forme abrégée ou avec le consentement du rédacteur. Le CNRC ne se tient en aucune façon responsable des opinions exprimées par un collaborateur ou des déclarations qui pourrait faire ce dernier. La publication d'annonces publicitaires, d'avis et autres n'implique pas l'endossement de leur contenu par les directeurs du *Journal* ou du CNRC. L'utilisation des noms de marques de commerce ne signifie aucunement une sanction du produit en question ou de la compagnie qui le fabrique.

La correspondance ayant trait aux manuscrits acceptés et les **épreuves corrigés** doivent être envoyées aux Presses scientifiques du CNRC, Conseil national de recherches du Canada, Ottawa, ON K1A 0R6, Canada.

Recommandations aux auteurs sont publiés une fois par année dans le premier numéro de chaque volume. Des copies, ainsi que la **Politique de publication**, sont disponibles auprès des Presses scientifiques du CNRC ou au site Web :
http://www.nrc.ca/cisti/journals/rj.html

Microfilms : On peut acheter sur microfilms de 16 ou 35 mm tous les numéros depuis 1934. Des photocopies d'articles ou de numéros individuels sont aussi en vente auprès de University Microfilms International, 330 North Zeeb Road, Ann Arbor, MI 48106, É.-U.

This journal is indexed or abstracted in / Ce journal est signalé ou résumé dans
Aquatic Sciences and Fisheries Abstracts, Biological & Agricultural Index, CAB Abstracts (Centre for Agriculture and Biosciences), Chemical Abstracts, Current Awareness in Biological Sciences, Current Contents, FAO Freshwater and Aquaculture Contents Tables, FAO Marine Science Contents Tables, GEOBASE – Geo Abstracts, Meteorological and Geoastrophysical Abstracts, Oceanic Abstracts, Science Citation Index, Biological Abstracts, Environment Abstracts, Environmental Periodicals Bibliography, GEOREF, Life Sciences Collection, and PASCAL Database

Volume 54, Number 2, February 1997

Canadian Journal of Fisheries and Aquatic Sciences

Published by the National Research Council of Canada

Volume 54, numéro 2, février 1997

Journal canadien des sciences halieutiques et aquatiques

Publié par le Conseil national de recherches du Canada

Editor / directeur scientifique D.G. Cook

Assistant to the Editor / adjointe au directeur scientifique **M.M. Milne**

Consulting Editor / rédacteur-conseil **P.E.K. Symons** (Victoria, B.C.)

Consulting Editor / rédactrice-conseil **N. Moir** (Nepean, Ont.)

Editorial Board / conseil de rédaction

S.E. Campana, *DFO/MPO, Dartmouth*
E.M.P. Chadwick, *DFO/MPO, Moncton*
L.M. Dickie, *DFO/MPO, Dartmouth*
G.S. Jamieson, *DFO/MPO, Nanaimo*
J.R.M. Kelso, *DFO/MPO, Sault Ste. Marie*
K.R. Munkittrick, *DoE/MdE, Burlington*
J.D. Neilson, *DFO/MPO, St. Andrews*
D.L.G. Noakes, University of Guelph
L.S. Parsons, *DFO/MPO, Ottawa*
R.M. Peterman, *Simon Fraser University*
R.D. Robarts, *DoE/MdE, Saskatoon*
J.C. Roff, *University of Guelph*
D.M. Rosenberg, *DFO/MPO, Winnipeg*
P. Schwinghamer, *DFO/MPO, St. John's*
(Deputy editor / rédacteur adjoint)
C.J. Walters, *University of British Columbia*
W.G. Warren, *DFO/MPO, St. John's*
N.H.F. Watson, *DFO/MPO, Halifax*

NRC Research Press / Presses scientifiques du CNRC

Editor-in-chief / directeur général **Bruce P. Dancik**

Assistant Editor-in-chief / directeur général adjoint **Iain E.P. Taylor**

Journals Program / Programme des revues

(Fax / télécopieur : 613-952-7656; e-mail / courrier électronique : research.journals@nrc.ca;
URL: http://www.nrc.ca/cisti/journals/)

Director / directrice **Aldyth Holmes**

Business Manager / gestionnaire des affaires **Paul McClymont**

Manager, Journals Program / gestionnaire, Programme des revues **Susan R. Strahlendorf**

Senior Publication Officer / agent principal de publication **Mike Boroczki**

Publication Officers / agentes de publication **Jennifer Douglas (613-993-0368), Jennifer McColl (613-993-4500)**

Publication Officer (French manuscripts) / agente de publication (manuscrits français) **Ginette Fortier (613-993-9117)**

Advertising / annonces publicitaires **Hoda Jabbour (613-993-9085)**

Reprints / tirés à part **Judy Heyman (613-993-0151)**

Subscriptions / abonnements **Helen Goulet (613-993-9084)**

© National Research Council of Canada 1997. World rights reserved.

No part of this publication may be reproduced, stored, or transmitted in any form or by any means, without the written permission of the publisher, except as stated below.

The National Research Council of Canada grants **permission*** to individuals to make single copies of articles for personal use. Any person may reproduce short excerpts from articles in the journal for any purpose that respects the moral rights of authors, provided that the source is fully acknowledged. As a courtesy, the consent of authors of such material should be obtained directly from the author.

Authorization to reproduce items for other than personal use, as stated above, is granted by the National Research Council of Canada provided that the copyright fee of $7.00 per copy is paid directly to the Copyright Clearance Center (CCC) Transactional Reporting Service, 21 Congress Street, Salem, MA 01970, U.S.A. 0706-652x/97 $7.00 + 0.00.

*The above permission does not extend to other kinds of copying, such as copying for general distribution, for advertising, or promotional purposes, for creating new collective works, or for resale. For such copying, arrangements must be made with the publisher, National Research Council of Canada, Ottawa, ON K1A 0R6, Canada.

© Conseil national de recherches du Canada 1997. Tous droits réservés.

Il est interdit de reproduire, de sauvegarder ou de transmettre quelque partie de la présente publication dans quelque format ou de quelque façon sans le consentement écrit de l'éditeur, à l'exception des conditions suivantes.

Le Conseil national de recherches du Canada accorde aux particuliers le droit de faire des copies individuelles pour leur usage personnel. Toute personne peut reproduire de courts extraits des articles de la Revue pour autant que les droits moraux de l'auteur soient respectés et que la mention de provenance soit clairement indiquée. Il est de convenance de demander le consentement de l'auteur avant de faire mention de son travail.

On obtiendra l'autorisation de reproduire des articles à d'autres fins que l'usage personnel (tel qu'on l'a décrit ci-dessus) auprès du Conseil national de recherches du Canada pour autant que des redevances de droits d'auteurs de 7,00 $/copie soient versées au Copyright Clearance Center (CCC) Transactional Reporting Service, 21 Congress Street, Salem, MA 01970, U.S.A. 0706-652x/97 7,00 $ + 0,00.

*La permission ci-dessus ne s'applique pas à la reproduction à grande échelle, telle que la reproduction aux fins de distribution générale, publicitaire ou promotionnelle, de compilation de nouveaux ouvrages collectifs ou de revente. Dans de tels cas, il faudra ratifier une entente particulière avec l'éditeur, le Conseil national de recherches du Canada, Ottawa, ON K1A 0R6, Canada.

Subscription information

The *Canadian Journal of Fisheries and Aquatic Sciences* (ISSN 0706-652X) is published monthly by the National Research Council of Canada. 1997 annual subscription rates (second class postage) for Canada are $461CAN institutional and $157CAN personal, for US & foreign $461US institutional and $167US personal. For air mail add $150US to foreign rate. Canadian customers please include 7% GST (business no. 12149 1807 RT 0275). In Quebec please add a further 6.5% QST to the total price including GST (QST no. 1006178088). Subscriptions are entered for the calendar year. Remittances should be made payable to the Receiver General for Canada credit NRC. Price for regular single issues: institutional, $60.00; personal, $25.00. Supplements will be priced separately. Special rates apply to members of Canadian scientific and engineering societies if they subscribe through their respective societies.

There are no refunds on cancelled subscriptions once delivery has begun. Claims for missing issues should be made within four months (N. America) or six months (outside N. America) following the date of publication. The publisher expects to supply missing issues only when losses have occurred in transit and when the reserve stock will permit. For change of address and subscription enquiries: Subscription Office, NRC Research Press, National Research Council Canada, Ottawa, ON K1A 0R6, Canada; tel: 613-993-9084; fax: 613-952-7656; e-mail: research.journals@nrc.ca.

Microform backfiles are available from UMI, P.O. Box 1346, Ann Arbor, MI 48106-1346, U.S.A., and Micromedia Ltd., 20 Victoria St., Toronto, ON M5C 2N8, Canada.

Renseignements sur l'abonnement

Journal canadien des sciences halieutiques et aquatiques (ISSN 0706-652X) est une publication mensuelle du Conseil national de recherches du Canada. Le prix de l'abonnement annuel au Canada (courrier de deuxième classe) est de 461 $CAN pour les organismes et de 157 $CAN pour les particuliers. À l'étranger (y compris les É.-U.), le prix de l'abonnement est de 461 $US pour les organismes et de 167 $US pour les particuliers. L'expédition par avion exige des frais de 150 $US en sus du prix d'abonnement à l'étranger. Les clients du Canada doivent ajouter la TPS de 7% (numéro d'entreprise 12149 1807 RT 0275). Au Québec, il faut ajouter la TVQ de 6,5% au prix total, TPS comprise (nº de TVQ 1006178088). Les abonnements sont pour l'année civile. Tout paiement doit être adressé au Receveur général du Canada, crédit du CNRC. Le prix par numéro est de 60 $ pour les organismes et de 25 $ pour les particuliers. Le prix des hors-série varie. Les membres des sociétés canadiennes de sciences et de génie bénéficient de taux préférentiels en s'abonnant via leurs sociétés respectives.

Aucun remboursement ne sera accordé ni aucune annulation acceptée une fois que les livraisons auront commencé. Les réclamations de numéros manquants devront être soumises dans les quatre mois (Amérique du Nord) ou dans les six mois (à l'extérieur de l'Amérique du Nord) suivant la date de publication. L'éditeur ne fournira de numéros manquants que si le document a été perdu pendant l'expédition et que si le stock le permet. Adressez tout changement d'adresse ou toute question quant à l'abonnement au : Bureau des abonnements, Presses scientifiques du CNRC, Conseil national de recherches Canada, Ottawa, ON K1A 0R6, Canada; téléphone : 613-993-9084; télécopieur : 613-952-7656; courrier électronique : research.journals@nrc.ca.

On peut obtenir les fichiers rétrospectifs sur **microforme** auprès de UMI, P.O. Box 1346, Ann Arbor, MI 48106-1346, É.-U., et auprès de Micromedia Ltée, 20 rue Victoria, Toronto, ON M5C 2N8, Canada.

ANNOUNCEMENT / ANNONCE

After a 22-year association with the *Journal*, David Cook is retiring as Editor, a position he has held for the last 9 years. NRC is pleased to welcome Drs. John Roff and Moira Ferguson (University of Guelph) as the new Co-editors, to whom all new submissions should be directed as of 1 June 1997. Dr. Cook will continue to handle work in progress until 1 September 1997.

The 3-month transition period will minimize disruption for authors and readers. It will give the new Editors time to recruit their Editorial Board and establish editorial procedures. I am confident that with the continued support of the community, the new Editors and their Board will maintain the *Journal*'s high standards and well-deserved reputation for publishing the very best research.

I would like to acknowledge the debt that is owed to Dr. Cook for his commitment and service to the *Journal* and the aquatic sciences community that it serves. His wise and careful guidance of the *Journal* was instrumental in its continuing quality and success. David's standards of editorial integrity are a model for any journal editor.

Après 22 ans de loyaux services pour le *Journal*, David Cook a décidé d'abandonner les rênes de la direction scientifique, qu'il tenait depuis 9 ans. Le CNRC est heureux d'annoncer la nomination de M. John Roff et de M^{me} Moira Ferguson (University of Guelph) à titre de codirecteurs scientifiques du *Journal*. C'est à eux que devront être adressées les soumissions d'article à partir du 1er juin 1997. M. Cook assurera l'achèvement des projets entrepris sous sa gouverne et nous quittera officiellement le 1er septembre 1997.

Cette période de chevauchement de 3 mois vise à assurer une transition sans accroc pour les auteurs et les lecteurs. Elle permettra aux nouveaux directeurs scientifiques de mettre sur pied leur nouvelle équipe de rédaction et d'établir les procédures éditoriales pertinentes. Je suis certain qu'avec l'appui de la communauté, les nouveaux directeurs scientifiques et le nouveau comité de rédaction maintiendront les normes élevées de qualité du *Journal* et son enviable réputation en tant que véhicule de recherche supérieure.

J'aimerais exprimer ma sincère gratitude à M. Cook pour son engagement à l'égard du *Journal* et de la communauté des sciences aquatiques qu'il dessert. Son expérience et son savoir-faire ont été des éléments cruciaux de la qualité et du succès du *Journal*. Les normes d'intégrité en matière de publication de David sont un exemple qui devrait inspirer tous les directeurs scientifiques.

Dr. Bruce P. Dancik

Editor in Chief / Directeur scientifique en chef

Laboratory behavior of adolescent and adult males of the snow crab (*Chionoecetes opilio*) (Brachyura: Majidae) mated noncompetitively and competitively with primiparous females

Bernard Sainte-Marie, Jean-Marie Sévigny, and Yves Gauthier

Abstract: In the laboratory, virgin females of snow crab (*Chionoecetes opilio*) were placed singly with one or two males to investigate mating behavior. Physiologically mature males, both adolescent (i.e., chelae undifferentiated) and adult (i.e., chelae differentiated), mated with females. Typically, matings comprised a precopulatory period, one or more copulations, and a postcopulatory period during which the female extruded eggs. Some females copulated after oviposition. In noncompetitive mating, adult males of 40–60 mm carapace width had repeated but brief copulations, while adult males of 120–140 mm carapace width had one long copulation. In pairs of competing adult males, large body size was an advantage both in taking over a female from a rival and in preventing female takeover by a rival. However, in competitions between an adult male and a larger adolescent male, the adult was more likely to dispossess his rival or prevent takeover. The variance in mass of ejaculate stored by females was explained (98%) by total copulatory time and number of copulations. Females with two mates obtained more copulations and larger ejaculate stores than females with one mate, suggesting that interannual variability in ejaculate stores reported for wild females is related to the level of male sexual competition.

Résumé : Au laboratoire, des femelles vierges du crabe des neiges (*Chionoecetes opilio*) ont été placées individuellement avec un ou deux mâles afin d'étudier le comportement d'accouplement. Les mâles physiologiquement matures, adolescents (i.e., pinces non différenciées) et adultes (i.e., pinces différenciées), se sont accouplés avec les femelles. Les accouplements comprenaient normalement une période précopulatoire, une ou plusieurs copulations et une période postcopulatoire au cours de laquelle la femelle pondait. Certaines femelles ont copulé après la ponte. En situation non compétitive, les mâles adultes de 40–60 mm de largeur de carapace ont eu plusieurs épisodes brefs de copulation, alors que les mâles adultes de 120–140 mm de largeur de carapace ont eu une seule longue copulation. Lorsque deux mâles adultes étaient en compétition, une plus grande taille était avantageuse tant pour ravir la femelle au rival que pour empêcher le rival de s'approprier la femelle. Cependant, dans les compétitions opposant un mâle adulte à un mâle adolescent plus grand, l'adulte était plus susceptible de déposséder son rival ou de prévenir l'appropriation. La variance dans la masse des éjaculats entreposés par les femelles s'expliquait à 98% par le nombre et la durée totale des copulations. Les femelles ayant eu deux partenaires sexuels ont obtenus plus de copulations et d'éjaculat que les femelles n'ayant eu qu'un partenaire, ce qui suggère qu'une des causes de la variabilité interannuelle des réserves en éjaculat observée chez les femelles sauvages est l'intensité de la compétition sexuelle entre mâles.

Introduction

Male snow crab (*Chionoecetes opilio*) develop primary and secondary sexual characters in three stages (Comeau and Conan 1992; Sainte-Marie et al. 1995). The transition from one developmental stage to the next occurs at a critical molt, and males in either developmental stage can be recognized on the basis of chela allometry and physiological characteristics. The first critical molt marks the onset of spermatogenesis (i.e., physiological maturity) and occurs over a narrow range of body sizes. In recent years on the east coast of Canada, no examined male with carapace width (CW) <31 mm and all examined males with CW >40 mm had differentiated reproductive tracts that contained spermatophores. The second critical molt is a terminal molt, during which the chelae become differentiated, occurring over a broad range of body sizes. Terminally molted (i.e., adult) males may vary in CW from 40 mm (Sainte-Marie and Hazel 1992) to at least 155 mm (Bailey and Elner 1989), while nonterminally molted but sperm-producing (i.e., adolescent) males may reach a CW of at least 120 mm (Conan and Comeau 1986).

Field studies suggest a distinct mating hierarchy for physiologically mature males of *Chionoecetes opilio* on the basis of their developmental stage and size. Among hard-shelled males, adults may outcompete adolescents and large adults may outcompete small adults (Ennis et al. 1990; Comeau et al. 1991; Sainte-Marie and Hazel 1992; Elner and Beninger

Received January 30, 1996. Accepted July 16, 1996.
J13280

B. Sainte-Marie,[1] J.-M. Sévigny, and Y. Gauthier. Division des invertébrés et de la biologie expérimentale, Institut Maurice-Lamontagne, ministère des Pêches et des Océans, 850 route de la Mer, C.P. 1000, Mont-Joli, QC G5H 3Z4, Canada.

[1] Author to whom all correspondence should be addressed.
e-mail: b_stemarie@qc.dfo.ca

© 1997 NRC Canada

1995). In eastern Canada, the male-only crab fishery may have increased mating opportunities for adult males smaller than legal size (i.e., 95 mm CW) and for adolescent males of various sizes by reducing abundance of legal-sized adult males and thereby relaxing male sexual competition (Bailey and Elner 1989; Ennis et al. 1990; Sainte-Marie 1993). Biologists are concerned that shifts in the characteristics, and decreases in the abundance, of males available to breed females might have negative effects on population reproductive potential (e.g., Paul 1984; Bailey and Elner 1989; Kruse 1993).

Female *Chionoecetes opilio* have spermathecae to store sperm over extended periods of time for subsequent use in fertilizing egg clutches (Watson 1970; Beninger et al. 1988). In the laboratory the quantity of ejaculate and number of sperm received by virgin females from individual males are variable, but on average, adult males, irrespective of their size, along with adolescent males of 80–100 mm CW, may deliver enough sperm to fertilize at least two successive egg clutches produced 1 year apart (Sainte-Marie and Lovrich 1994; Sainte-Marie and Carrière 1995). Nevertheless, in nature, there is evidence that the quantity of sperm stored by females may vary interannually (Beninger et al. 1988; B. Sainte-Marie, unpublished data). To date, there is little information on the causes of variability in the quantity of ejaculate or sperm acquired by females, although Sainte-Marie and Lovrich (1994) suggested that it might depend on frequency and duration of copulation (i.e., period of mating when male gonopod is inserted in female vagina). In other crab species, male density and operational sex ratio (term defined by Emlen and Oring 1977) can affect male copulating frequency or duration of copulations and, consequently, the quantity of sperm passed to females (Wilber 1989; Jivoff 1995). Sperm stored over from the first (i.e., primiparous) mating period of female *Chionoecetes opilio* may be important for reproduction when or where males competent to mate multiparous females become limiting, owing to (*i*) high fishing mortality on large adult males (Comeau and Conan 1992; Sainte-Marie 1993) and (or) (*ii*) recurrent changes in the abundance, size structure, and relative proportions of adolescent and adult males in populations (Elner and Beninger 1995; Sainte-Marie et al. 1996). Thus, detailed knowledge of the mating behavior of males with primiparous females, and of the extent to which behavior can vary with male developmental stage and size, represents a potentially important contribution to the reproductive biology of this species. Research on primiparous females is further justified by the fact that they are an important component of reproduction (Sainte-Marie 1993), in some years contributing the majority of eggs produced by populations (Elner and Beninger 1995; Sainte-Marie et al. 1996).

Current knowledge of the primiparous mating behavior for *Chionoecetes opilio* is largely based on detailed and continuous observations of one mating pair (disturbed by observer) in Watson (1970), of two mating pairs in Watson (1972), and of three mating pairs (same male, three different females) in Takeshita and Matsuura (1980). Additionally, Watson (1972) casually observed a total of eight other mating pairs, involving only four different males. Reports concurred in showing that males and females had only one copulation lasting 41–45 min, but differed in the fact that egg extrusion occurred before (Takeshita and Matsuura 1980) or after (Watson 1972) mates parted. Males that mated females in these studies were of unknown developmental stage and represented a relatively narrow range of body sizes (60.8–98.2 mm CW). These caveats could be important because mating behavior can vary with male size in the closely related tanner crab (*Chionoecetes bairdi*) (Donaldson and Adams 1989) and depends on both male developmental stage and size in another majid crab, *Libinia emarginata* (Laufer and Ahl 1995). Preliminary observations on *Chionoecetes opilio* primiparous mating in Sainte-Marie and Lovrich (1994) indicated that more than one copulation could occur before mates parted and that the duration of copulation was far more variable (1–103 min) than previously reported. Other studies of primiparous or multiparous mating in *Chionoecetes opilio* rest on discontinuous or low-detail laboratory observations (Moriyasu and Conan 1988; Ennis et al. 1988; Claxton et al. 1994) or on field collections of mating pairs (Taylor et al. 1985; Conan and Comeau 1986; Hooper 1986; Ennis et al. 1988, 1990; Comeau et al. 1991; Sainte-Marie and Hazel 1992) and contain no information on frequency or duration of copulations.

This laboratory study provides quantitative information on the behavior of *Chionoecetes opilio* during primiparous mating, both with and without male sexual competition. The specific objectives were to (*i*) assess mating behavior in relation to differences in male developmental stage and body size and (*ii*) identify the proximate sources of variability in ejaculate delivery.

Materials and methods

Collection and holding of crabs

Chionoecetes opilio were collected in September–October of 1990, 1991, and 1992 from the Estuary and Gulf of the Saint Lawrence River, eastern Canada, and used during the following winter of each year. The width of the carapace at its broadest point excluding spines was measured to the nearest 0.1 mm on females and males using a standard vernier caliper (Sainte-Marie and Hazel 1992). For males, chela height excluding spines (CH) was determined to the nearest 0.1 mm at the highest point of the right propodus in a perpendicular plane to the long axis of the chela. Only males and immature females >40 mm CW were kept for the laboratory study. Males were classified as adolescent or adult by comparing their individual CH and CW measurements with a site-appropriate scatterplot and discriminant function for CH and CW (Sainte-Marie and Hazel 1992). The exoskeleton condition of males, which is an approximate index of time elapsed since the last molt, was determined following the criteria in Sainte-Marie et al. (1995).

Crabs were held in laboratory tanks supplied with fresh flowing seawater at monthly average temperatures of –0.4 to 3.7°C and salinities of 28–31‰. Photoperiod was controlled to reflect natural light rhythms. Females and males were kept in separate tanks with independent water supplies, and within tanks, males were further segregated by maturity and CW using dividers. Crabs were fed previously frozen shrimp (*Pandalus borealis*) or Atlantic herring (*Clupea harengus*) semiweekly. Tanks were checked for molting crabs daily from September to December and at least twice daily from January to March.

Mating experiments

Noncompetitive and competitive mating experiments were conducted. In noncompetitive mating experiments, one female was introduced into a tank containing one adult male in either of three size categories, 40–60, 80–100, or 120–140 mm CW, or one adolescent male of 80–100 mm CW. Noncompetitive mating experiments were conducted from 31 January to 2 March 1991 and from 21 January to 1

© 1997 NRC Canada

March 1992. In competitive mating experiments, one female was placed in a tank with two males. Pairs of competing males consisted of either an adult of 72.1–92.1 mm CW versus an adolescent of 84.5–102.3 mm CW, with a size difference of 7.9–25.9 mm CW in favor of the adolescent, or an adult of 70–94.8 mm CW versus an adult of 100.2–126.2 mm CW, resulting in a size difference of 22.3–52.1 mm CW in favor of the largest adult. In adult–adult competitions the male with the largest carapace also had the largest chelae, but in 6 of 12 adolescent–adult competitions the adolescent male, despite being larger in body size, had the smallest chelae (difference of 0.3–3.8 mm CH). Competitive mating experiments were conducted from 10 February to 25 March 1993.

Females in noncompetitive and competitive mating experiments were virgin and either had begun their maturity molt or had molted to maturity ≤12 h before introduction to a male. Males used in either type of mating experiment had two chelae. All adolescent males had exoskeletons in the intermediate condition (i.e., hard shell with light epibiont fouling) at the time of the mating experiment, but they subsequently molted <3 weeks or 12–14 months after mating. Exoskeletons of adult males were either in an intermediate or dirty–hard (i.e., dull hard shell with scars and usually heavy epibiont fouling) condition. Adult males used herein probably were in prime reproductive condition, on the basis of evidence for *Chionoecetes bairdi* indicating that males with either clean–hard or dirty–soft (i.e., decaying) exoskeletons are less competitive than males with intermediate or dirty–hard exoskeletons (Stevens et al. 1993; Paul et al. 1995). Males were acclimated to mating tanks for at least 6 h prior to experimentation.

Mating experiments were conducted in aquaria or tanks that had coarse sand substrate and running fresh seawater at −0.5 to 0.5°C. A 120-L aquarium was used for noncompetitive mating experiments. A mirror was set above the aquarium at an angle of approximately 45° with the water surface to provide a side-mounted, low-light video camera (Panasonic Lunar Lite) with simultaneous lateral and top views of the mating pair. For competitive mating experiments, 1000-L tanks with a diameter of 1.3 m were used and the same low-light video camera was mounted above the tank. Dim white (60 lx at water surface) and red fluorescent lighting was used during the diurnal and nocturnal phases of the experiments, respectively. All experiments were recorded on a time-lapse VCR recorder (Panasonic AG-6720A) at one image per second. Noncompetitive experiments were terminated either when the female had extruded eggs or 24 h after the female was placed with the male; competitive mating experiments ran for 24 h. Timing of female maturity molt, temperature, salinity, substrate, and size range of males in the laboratory mating experiments were similar to conditions for primiparous mating in nature (Sainte-Marie and Hazel 1992; Lovrich et al. 1995).

Depending on the type of experiment, video recordings were analyzed to extract some or all of the following data: time between female introduction or molt and start of first copulation (i.e., precopulatory period), number and duration of clasping bouts and identity of male clasper, number and duration of copulations and identity of copulating male, and number and duration of agonistic bouts between males and identity of the male initiating the bout. Agonistic interactions between competing males were classified in two broad categories: probing (i.e., brief contacts followed by retreat) and attacking (i.e., lunging toward opponent, grasping opponent or female held by opponent, fighting). Some interactions occurred between males before the female was introduced, but they were of low intensity and consisted mostly of probing.

Females used in noncompetitive matings were kept alive and monitored for egg development as described in Sainte-Marie and Carrière (1995). Females used in competitive matings were killed immediately after the experiment ended and the quantity of ejaculate in their spermathecae was measured as blotted mass with the lower limit for reliable measurement being 10^{-3} g (Sainte-Marie and Lovrich 1994). The quantity of stored ejaculate in individual females is presented as the mean of ejaculate mass in the right and left spermathecae. Ejaculate mass may be a reasonably good proxy of the number of sperm in storage (Sainte-Marie and Lovrich 1994) when determined at a constant time after insemination.

Statistics presented here for any variable are the mean and standard error. Student's t test was used to compare means between two samples, while the nonparametric Kruskal–Wallis test was used to compare means for double or multiple sets of data that were not homoscedastic. Correlation and simple or multiple linear regression were used to determine relationships between ejaculate mass, number of copulations, and (or) total copulatory time for females. Fisher's exact test was used to test for differences between competing categories of males in success rate of attacks for appropriation of a female, and the G test for goodness of fit was used to test if competing categories of males were equally successful in obtaining first and last copulations with females.

Results

Noncompetitive mating experiments

Of the 45 females that mated noncompetitively, none lost pereiopods, and only three (6.7%) failed to copulate and did not extrude eggs while in the presence of a male for 24 h (Fig. 1). For each failure, the male was an adult of 120–140 mm CW.

Males usually clasped females within minutes of their introduction into the aquarium and then held them in a rostrum to rostrum position or, infrequently, stood over the female until copulation began. The mean precopulatory period did not vary significantly among size-classes of adult males, but was significantly longer for adolescent than for adult males of 80–100 mm CW (Table 1). Adult males of 40–60 mm CW had difficulty positioning larger females for copulation, and in two cases, copulation occurred with the male lying on his back and the female above. Adult males of 120–140 mm CW did not always respond to females by initially clasping and guarding them: in four cases the female positioned herself for copulation alone, with the male intervening only to lower his abdomen, and subsequently the female clung to the ventral side of the male.

During copulation, the male's gonopods were only inserted shallowly into the female's gonopores, and no flexing of female or male abdomens was perceived. However, the pumping action of the male's second gonopod within the first gonopod was obvious on a few occasions. The first copulation lasted significantly longer for adult males of 120–140 mm CW than for adult males of 40–60 or 80–100 mm CW (Table 1). The mean number of copulations over 24 h was inversely related to adult male CW, ranging from 1 for males of 120–140 mm CW to 2.8 for males of 40–60 mm CW (Table 1). However, mean total copulation time did not vary significantly among the size categories of adult males (Table 1). The mean duration of first copulation did not differ significantly between adolescent and adult males of 80–100 mm CW, but data for 24-h matings were too few to compare mean number of copulations and mean total copulation time between the two categories of males (Table 1).

Females extruded eggs while held by the male during a postcopulatory guarding period that lasted 4–6 h. Among the nine females that copulated more than once, egg extrusion occurred after the first (eight of nine cases) or the second (one case) copulation. After extruding eggs, the female was released by the male and subsequent interactions usually

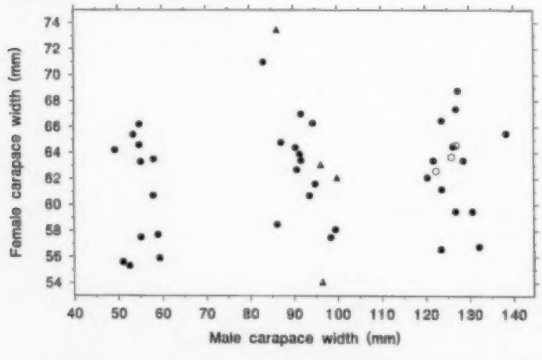
© 1997 NRC Canada

Table 1. Duration of the precopulatory period, first copulation, and all copulations, and the number of copulations for adult and adolescent males of various CW size-classes in noncompetitive mating with female *Chionoecetes opilio*.

	Adult (40–60 mm CW)	Adult (80–100 mm CW)	Adult (120–140 mm CW)	Adolescent (80–100 mm CW)
Precopulatory period (min)				
Mean ± SE	131.9±30.9	90.3±31.1	179.8±44.8	403.4±198.8
Range	13.4–370.9	6.6–401.2	3.1–601.4	69.6–968.8
N	12	13	13	4
H			3.78	4.63
P			0.151	0.032
First copulation (min)				
Mean ± SE	14.8±2.2	14.3±1.2	23.5±5.1	22.6±8.2
Range	9.8–37.1	6.9–22.0	12.3–80.3	10.4–46.7
N	12	13	13	4
H			6.53	0.82
P			0.038	0.365
Number of copulations				
Mean ± SE	2.8±0.5	1.6±0.6	1.0±0.0	1.0±0.0
Range	1–7	1–4	1	1
N	12	5	8	2
H			8.56	
P			0.014	
All copulations (min)				
Mean ± SE	45.4±13.2	33.4±22.0	19.3±3.0	14.1±3.7
Range	12.8–177.9	6.9–121.0	12.3–35.9	10.4–17.8
N	12	5	8	2
H			4.51	
P			0.105	

Note: The Kruskal–Wallis test (*H* statistic) compared differences among size-classes of adult males and between adolescent and adult males of 80–100 mm CW. Statistics on the number of copulations and the duration of all copulations are based only on experiments that lasted 24 h.

Fig. 1. CW of male and postmolt female *Chionoecetes opilio* in noncompetitive mating experiments. Circles and triangles represent matings with adult and adolescent males, respectively. Solid symbols indicate that mating resulted in production of an egg clutch; open symbols indicate that the female did not extrude eggs while in the presence of a male for 24 h.



consisted of brief episodes (<10 min) of probing and clasping by the male. When additional copulations occurred, the berried female was usually freed minutes after copulation. The duration of copulations before (15.9 ± 8.8 min) and after (18.1 ± 8.0 min) egg extrusion did not differ (*t* test: $t = -0.54$, $P = 0.60$) for females that copulated in both conditions. One female copulated five times after extruding eggs. Thirty-eight of the 42 females that copulated eventually hatched their first clutch of eggs.

Competitive mating experiments

All of the 24 competitively mated females copulated at least once. Only two (8.3%) females failed to extrude their eggs while in the presence of males for 24 h (Fig. 2). Both failures involved females that copulated with only one of the competing males. Seven females lost one to four pereiopods each during fights between competing males. Only one male suffered visible injury in fighting, losing one pereiopod.

When adult males competed with larger adolescent males, the adults clasped the female on average four times longer than the adolescents (Table 2). Both adolescent and adult males probed and attacked their opponent while he clasped a female. However, on average, adolescent males probed 0.2 times and attacked 0.3 times per 100 min of opponent clasping time, while adult males probed 0.4 times and attacked 0.9 times per 100 min of opponent clasping time. Moreover, attacks by adult males were more likely to result in a female takeover than were attacks by adolescent males (Table 2). The only adolescent males that were successful in

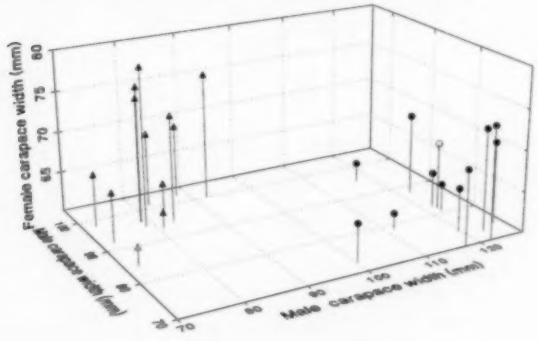
© 1997 NRC Canada

Table 2. Mean and standard error of clasping time and number and duration of agonistic bouts and percentage of successful attacks by pairs of males competing for female *Chionoecetes opilio*.

	Adult × adolescent		Adult × adult	
	75–92 mm	84–102 mm	100–126 mm	70–95 mm
Clasping female (min)	649.6±202.2	157.1±60.3	367.5±88.4	207.2±71.2
Probing opponent (min)	0.7±0.3	0.9±0.4	0.3±0.2	1.0±0.4
Number of probes	0.7±0.3	1.6±0.8	0.4±0.2	1.9±0.6
Attacking opponent (min)	3.6±2.4	3.8±1.4	2.9±1.0	5.2±2.5
Number of attacks	1.4±0.5	1.8±0.5	1.9±0.6	3.3±1.4
% successful attacks	83.3	30.8	100.0	11.1

Note: Competitions were one adult male of 72–92 mm CW against one adolescent male of 84–102 mm CW (N = 12) or one adult male of 100–126 mm CW against one adult male of 70–95 mm CW (N = 12). Agonistic behaviors consisted of probing or attacking the opponent while he clasped the female. Successful attacks resulted in female takeover. Differences in clasping time were tested with the Kruskal–Wallis test (H statistic): H = 6.75, P = 0.009 for adult versus adolescent, and H = 1.87, P = 0.172 for adult versus adult. Success rates of attacking males were compared using Fisher's exact test: P = 0.057 for adult versus adolescent and P < 0.001 for adult versus adult.

Fig. 2. CW of competing males and postmolt female *Chionoecetes opilio* in competitive mating experiments. Circles and triangles represent competition involving adult–adult or adult–adolescent males, respectively. Solid symbols indicate that mating resulted in production of an egg clutch; open symbols indicate that the female did not extrude eggs while in the presence of a male for 24 h.



taking over a female from an adult male were those with larger chelae than their opponent. Adult males copulated at least once with the female in 11 of 12 experiments, while the adolescent male was able to copulate in only half of the experiments (Table 3). Almost equal numbers of adult and adolescent males were responsible for the females' first copulation (Table 3). However, over time, adult males displaced adolescent males and ensured a significantly greater proportion of the females' last copulation (Table 3). Overall, adult males obtained more copulations and copulated longer than adolescent males (Table 3).

When a large adult male competed with a smaller adult male, there was no significant difference in mean clasping time by the two categories of males (Table 2). Smaller adult males probed their opponent twice as often as large adult males (0.5 versus 0.2 probes per 100 min of opponent clasping time), but the two size categories attacked their opponent with equal frequency (0.9 attacks per 100 min of opponent clasping time). However, attacks by large adult males consistently resulted in takeover of the female, while smaller adult males rarely won (Table 2). Large adult males copulated at least once with the female in 8 of 12 experiments, while smaller adult males copulated in 7 of the experiments (Table 4). Large and smaller adult males were responsible for almost equal numbers of the females' first copulation and for equal numbers of the females' last copulation (Table 4). The mean number of copulations and total copulation time did not differ significantly between large and smaller adult males (Table 4). On average, smaller adult males obtained two times fewer copulations in the presence of large adult males than in the presence of adolescent males (Tables 3 and 4).

The ceiling-mounted camera did not offer a view that allowed us to determine when females extruded eggs. However, in each competitive mating experiment that ended with the female carrying eggs, there occurred a conspicuously long postcopulatory guarding period (Tables 3 and 4) that lasted 5.5 ± 0.4 h.

There were no differences between females mated in adult–adolescent versus adult–adult competitions either in the mean duration of precopulatory period (187.7 versus 117.7 min, Kruskal–Wallis test, H = 0.12, P = 0.729), mean number of copulations (2.8 versus 2.0, H = 2.06, P = 0.151), mean total copulatory time (47.6 versus 42.0 min, H = 0.48, P = 0.488), or mean quantity of stored ejaculate (0.122 versus 0.124 g per spermatheca, H = 0.16, P = 0.686). However, Table 5 shows that females that copulated with both mates (i.e., biandrous) obtained significantly more copulations, longer total copulatory times, and larger stores of ejaculate than females that copulated with only one mate (i.e., monandrous).

Ejaculate delivery in competitively mated females

None of the examined spermathecae were filled to capacity. Overall, the mass of ejaculate stored in competitively mated females was positively correlated with the number of copulations (r = 0.46, P = 0.023) and total copulatory time (r = 0.87, P < 0.001). However, two outliers were obvious in the

© 1997 NRC Canada

Table 3. Duration of first copulation and of all copulations, number of copulations, and percentage of first and last copulations for adult males of 72–92 mm CW (adu) and adolescent males of 84–102 mm CW (ADO) competing in pairs for one female *Chionoecetes opilio* ($N = 12$).

	Adult, 72–92 mm	Adolescent, 84–102 mm	Test		
			H	G_{adj}	P
First copulation (min)	15.4±1.9 (8.1–23.4)	16.0±2.1 (11.9–23.6)	0.06		0.808
Number of copulations	2.1±0.4 (0–5)	0.8±0.3 (0–2)	5.84		0.016
All copulations (min)	35.4±7.2 (0–67.7)	12.2±4.1 (0–37.6)	4.93		0.026
% first copulation	58	42		0.32	>0.50
% last copulation	83	17		5.59	<0.05

Note: Values are given as the mean ± SE, with range in parentheses. The Kruskal–Wallis test (H statistic) compared differences in means, and the G test for goodness of fit was used to test the hypothesis that competing males obtained 50% of copulations. Actual copulating sequences for males in individual experiments (P is a protracted postcopulatory guarding period) were as follows: adu–P ($N = 2$); adu–P–adu; adu–adu–P; adu–adu–adu; adu–P–adu–adu–adu; adu–ADO–P–adu–adu–adu–adu; ADO–P–adu–adu–adu; ADO–adu–P; ADO–P–adu–ADO–adu; ADO–adu–P–ADO; ADO–P–ADO.

Table 4. Duration of first copulation and of all copulations, number of copulations, and percentage of first and last copulations for adult males of 100–126 mm CW (ADU) and adult males of 70–95 mm CW (adu) competing in pairs for one female *Chionoecetes opilio* ($N = 12$).

	Adult, 100–126 mm	Adolescent, 70–95 mm	Test		
			H	G_{adj}	P
First copulation (min)	20.7±5.7 (4.9–49.4)	14.6±0.8 (11.6–16.3)	0.16		0.685
Number of copulations	0.9±0.2 (0–2)	1.1±0.3 (0–3)	0.02		0.879
All copulations (min)	23.5±7.8 (0–91.4)	18.5±5.7 (0–59.8)	0.13		0.722
% first copulation	58	42		0.32	>0.50
% last copulation	50	50		0.00	>0.90

Note: Values are given as the mean ± SE, with range in parentheses. Other information as in Table 3. Copulating sequences: ADU; ADU–P ($N = 2$); ADU–P–ADU; ADU–ADU–P; ADU–P–adu–ADU; ADU–P–adu; adu–P–ADU–adu; adu–P; adu–P–adu; adu–P–adu–adu ($N = 2$).

scattergram of ejaculate mass on total copulatory time (Fig. 3). One monandrous female with three copulations contained no measurable amount of ejaculate, and one monandrous female with four copulations had spermathecae that were unequally loaded (0.141 g in the right versus <0.001 g in the left spermatheca). In both cases, the mates were adult and the ratio of female CW to male CW fell within the range of ratios for other mating pairs in which females stored larger masses of ejaculate.

Sources of variability in ejaculate mass were investigated by multiple regression, given the possibly confounding effects of number of copulations and total copulatory time that were strongly correlated ($r = 0.75$, $P < 0.001$). With the two outliers excluded, the mass of stored ejaculate in monandrous females varied as a function of both number of copulations ($P < 0.001$) and total copulatory time ($P < 0.001$) (Fig. 3). The negative partial regression coefficient for the number of copulations indicated that ejaculate mass decreased with increasing number of copulations for a constant total copulatory time. Insight into the nature of this decrease was obtained by examining the results of multiple regression of ejaculate mass (grams) on the duration of first (FT, minutes) and second (ST, minutes) copulations for monandrous females with only two copulations:

$$\text{ejaculate mass} = 0.0043\text{FT} + 0.0034\text{ST} - 0.0254,$$
$$R^2 = 0.995,\ P < 0.001,$$

in which both partial regression coefficients were significant (FT: $P = 0.001$; ST: $P = 0.004$). We infer from this analysis that the quantity of ejaculate passed to females for a given copulatory time was less during the second than during the first copulation.

By contrast, multiple regression showed for biandrous females that total copulatory time contributed significantly ($P < 0.001$) and number of copulations nonsignificantly ($P = 0.174$) to variation in mass of stored ejaculate (Fig. 3). However, for a constant total copulatory time, biandrous females had less stored ejaculate than any monandrous female except the two outliers (Fig. 3).

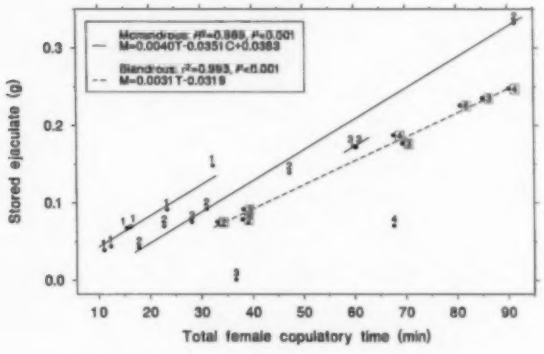
© 1997 NRC Canada

Table 5. Number of copulations, total copulatory time, and mass of stored ejaculate per spermatheca obtained by females from one (monandrous) or from both (biandrous) of two competing male *Chionoecetes opilio*.

			Test	
Female performance	Monandrous	Biandrous	*H*	*P*
Number of copulations	1.9±0.2	3.4±0.5	6.94	0.008
Total copulatory time (min)	36.5±5.7	62.8±8.2	6.61	0.010
Stored ejaculate mass (g)	0.105±0.020	0.165±0.026	4.59	0.032

Note: Values are given as the mean ± SE. Competitions were adult males of 72–92 mm CW against adolescent males of 84–102 mm CW or adult males of 100–126 mm CW against adult males of 70–95 mm CW. The Kruskal–Wallis test (*H* statistic) was used to compare performance of monandrous and biandrous females.

Fig. 3. Mean mass (M, grams) of ejaculate stored per spermatheca of competitively mated female *Chionoecetes opilio* in relation to total female copulatory time (T, minutes). Numbers beside data points represent the number of copulations (C) realized by each female. Unboxed numbers indicate that the female was monandrous (copulation with one mate); boxed numbers indicate that the female was biandrous (i.e., copulation with both mates). Monandrous females with either three copulations and <0.001 g of stored ejaculate or with four copulations and 0.07 g of stored ejaculate were excluded from the regression analyses.



Discussion

Mating behavior

The present observations establish the potential of female *Chionoecetes opilio* for repeated mating (i.e., more than one copulation with a single mate) and multiple mating (i.e., copulation with more than one mate), immediately after the maturity molt. Polyandry has been documented for other majids (Hinsch 1968; Diesel 1991) and for *Chionoecetes bairdi* in particular (Donaldson and Adams 1989). Among several proposed explanations for repeated or multiple mating in a variety of taxa (Parker 1970; Walker 1980; Drummond 1984; Halliday and Arnold 1987; Hunter et al. 1993), the necessity for virgin females to accumulate enough good sperm to ensure fertilization of the first and potentially of the subsequent egg clutches may be the most likely reason for *Chionoecetes opilio*.

Female *Chionoecetes opilio* can copulate while they carry an egg clutch, as seen also in the majids *Maja verrucosa* (Knudsen 1964), *L. emarginata* (Hinsch 1968), and *Pleistacantha moseleyi* (Berry and Hartnoll 1970). The presence of large, apparently fresh deposits of ejaculate in the spermathecae of female *Chionoecetes opilio* carrying embryos either halfway through development time (Sainte-Marie 1993) or fully developed (Taylor et al. 1985) had suggested the possibility of copulation some time between egg extrusion and larval release. The observation of alternating layers of eggs and ejaculate in the spermathecae of exceptional females further suggests that oviposition can be interrupted by copulation (Sainte-Marie and Lovrich 1994). Given that primiparous female *Chionoecetes* spp. can be polyandrous and that sperm from the last mate may have precedence in fertilizing eggs (Paul et al. 1983; Elner and Beninger 1992; Sévigny and Sainte-Marie 1996), copulation during oviposition implies the possibility of multiple paternity for first broods, while copulation shortly after oviposition implies the possibility of different paternity for the first and second egg clutch even in females that rely only on sperm stored over from the first mating period to produce the second egg clutch. Although eggs may only be loosely attached to female pleopods shortly after oviposition, no detectable egg loss resulted from postextrusion copulations, which is not inconsistent with the observation that both the female and male are apparently quiescent during the copulatory act (the present study).

In noncompetitive mating, egg extrusion generally occurred before mates parted. Postcopulatory guarding in crustacean decapods may serve to protect the soft female from predators (e.g., Hartnoll 1969; Atema et al. 1979) and (or) to assure the male that he is the sole male parent of a female's brood (Wilber 1989; Diesel 1991; Jivoff 1995). This last possibility may be the principal reason in *Chionoecetes opilio*, since (*i*) hard-shelled multiparous females also are guarded after copulation (Taylor et al. 1985; Hooper 1986; Moriyasu and Conan 1988; Claxton et al. 1994) and (*ii*) some females that had extruded eggs and were still soft from molting in the present experiments were inseminated but not guarded extensively by males. This latter situation bears some resemblance to the reproductive behavior of *Cancer gracilis* in which primary males copulate with soft-shelled mature females and subsequently guard them for extended periods of time, while secondary males may copulate with the same females but do not guard them (Orensanz et al. 1995).

© 1997 NRC Canada

The proximate sources of variation among females in the quantity of stored ejaculate were the duration of copulation and number of copulations (Fig. 3). Reduced ejaculate delivery during the second copulation of monandrous females probably reflected a decrease in the rate of ejaculate delivery, although we cannot rule out the possibility of prolonged copulation after complete delivery of ejaculate. Prolonged copulation with reduced or no ejaculate delivery is demonstrated for some insect species and represents a form of contact mate guarding (e.g., Parker 1970; Thornhill and Alcock 1983), and some indirect evidence suggests that it exists in other crab species as well (Elner et al. 1987). The finding that biandrous females stored less ejaculate than monandrous females for a constant total copulatory time appears to be consistent with the hypothesis that males can extract a competitor's reproductive products from the spermathecae to reduce sperm competition (Beninger et al. 1991; Elner and Beninger 1992). However, the extremely tight relationship between ejaculate mass and total copulatory time for biandrous females (r^2 = 0.993) and the fact that insertion of male gonopods into the vagina is in appearance only superficial could instead suggest that copulation is devoted exclusively to ejaculate delivery, albeit at an even lesser rate than observed during the second copulation of monandrous females. The observed variability in ejaculate delivery across successive copulations in *Chionoecetes opilio* challenges Sainte-Marie and Lovrich's (1994) assumption that the number of sperm delivered to a female is directly proportional to the number of copulations, along with their subsequent calculation of the number of sperm expended per egg during fertilization, which may be slightly overestimated.

Behavior of males in noncompetitive mating varied with developmental stage and body size. The precopulatory period was longer for adolescent than for adult males (Table 1 herein; Claxton et al. 1994), possibly owing to male inexperience or female resistance to copulation. As in *Chionoecetes bairdi* (Donaldson and Adams 1989), the smallest male *Chionoecetes opilio* tended to have several short copulations, while the largest males had only one protracted copulation (Table 1). These two contrasting behaviors are similar to the re-mater and resistor types seen in insect species with high levels of male sexual competition (Gromko et al. 1984; López-León et al. 1993). Resistor males acquire females, fend off rivals, and control females until they have extruded eggs to ensure that they are the sole male parent. Re-mater males, which can be displaced by more competitive males, have brief copulations but copulate more often to increase the probability of gaining paternity of a brood.

Male sexual competition was beneficial to females in providing them with larger stores of ejaculate. Biandrous females contained more ejaculate than competitively mated monandrous females (Table 5), which in turn had larger ejaculates than females mated noncompetitively with adult males in the same year and experimental setup (0.080 ± 0.018 g per spermatheca; data from Sainte-Marie and Lovrich 1994). This pattern is consistent with behavioral responses to male sexual competition reported for the stone crab (*Menippe* spp.) (Wilber 1989), the blue crab (*Callinectes sapidus*) (Jivoff 1995), and a variety of insect species (e.g., Smith 1980; Dickinson 1986). Male sexual competition could explain Sainte-Marie and Lovrich's (1994) observation that spermathecae were more loaded in wild primiparous females than in primiparous females mated noncompetitively in the laboratory. Thus, natural and pronounced cycles or fishery-induced changes in the operational sex ratio of *Chionoecetes opilio* (Sainte-Marie et al. 1996), which affect levels of male sexual competition, may be one ultimate reason for interannual variation in the quantity of ejaculate stored by primiparous females.

Significance of laboratory results to the field

This study has (*i*) established hitherto unknown mating behaviors for *Chionoecetes opilio*, (*ii*) demonstrated significant differences in mating behavior between males in relation to developmental stage and body size, and (*iii*) identified two proximate causes and suggested one ultimate cause of variability in the quantity of ejaculate stored by primiparous females. Nevertheless, our effort may offer only a glimpse of the intricacies of mating in nature. For example, our data suggest that if adolescent males have relatively larger chelae than adult males, they may successfully compete for females, while recent evidence shows for *Chionoecetes bairdi* that competitive mating ability of hard-shelled adult males also depends on time elapsed since last molt (Stevens et al. 1993; Paul et al. 1995).

A major difference between the present laboratory conditions and the field is that the imminent premolt or postmolt females used herein resulted in the precopulatory period lasting at most only several hours whereas in nature, semiochemicals emitted by pubescent females (Bouchard et al. 1996) initiate male precopulatory behavior up to 8–13 days prior to female maturity molt (Watson 1972; Sainte-Marie and Lovrich 1994). This laboratory artefact may have prevented interactions between competing males from unfolding completely and may have favored chance matings. Although laboratory observations may be subject to bias, they represent an essential step towards improving knowledge of *Chionoecetes opilio* mating behavior because ice cover and extreme cold during the primiparous mating period compromise the quality and quantity of information that can be derived from field investigations (Moriyasu and Conan 1988; Sainte-Marie and Hazel 1992).

Acknowledgements

F. Hazel, P. Goudreau, and M. Bélanger assisted in the field and laboratory. We thank S. Bouchard, F. Hazel, and G.A. Lovrich for discussion and M. Comeau, R.W. Elner, P.R. Jivoff, J.N. McNeil, and anonymous reviewers for their thoughtful comments on earlier versions of this paper. This work was supported by the Québec Federal Fisheries Development Program.

References

Atema, J.S., Jacobson, S., Karnofsky, E., Oleszko-Szuts, S., and Stein, L.S. 1979. Pair formation in the lobster, *Homarus americanus*: behavioral development, pheromones and mating. Mar. Behav. Physiol. **6**: 277–296.

Bailey, R.F.J., and Elner, R.W. 1989. Northwest Atlantic snow crab fisheries: lessons in research and management. *In* Marine invertebrate fisheries: their assessment and management. *Edited by* J.F. Caddy. John Wiley & Sons, New York. pp. 261–280.

Beninger, P.G., Elner, R.W., Foyle, T.P., and Odense, P.H. 1988. Functional anatomy of the male reproductive system and the

© 1997 NRC Canada

female spermatheca in the snow crab *Chionoecetes opilio* (O. Fabricius) (Decapoda: Majidae) and a hypothesis for fertilisation. J. Crustacean Biol. **8**: 322–332.

Beninger, P.G., Elner, R.W., and Poussart, Y. 1991. The gonopods of the majid crab *Chionoecetes opilio* (O. Fabricius). J. Crustacean Biol. **11**: 217–228.

Berry, P.F., and Hartnoll, R.G. 1970. Mating in captivity of the spider crab *Pleistacantha moseleyi* (Miers) (Decapoda, Majidae). Crustaceana (Leiden), **19**: 214–215.

Bouchard, S., Sainte-Marie, B., and McNeil, J.N. 1996. Indirect evidence indicates female semiochemicals release male precopulatory behaviour in the snow crab, *Chionoecetes opilio* (Brachyura: Majidae). Chemoecology, **7**: 39–44.

Claxton, W.T., Govind, C.K., and Elner, R.W. 1994. Chela function, morphometric maturity, and the mating embrace in male snow crab, *Chionoecetes opilio*. Can. J. Fish. Aquat. Sci. **51**: 1110–1118.

Comeau, M., and Conan, G.Y. 1992. Morphometry and gonad maturity of male snow crab, *Chionoecetes opilio*. Can. J. Fish. Aquat. Sci. **49**: 2460–2468.

Comeau, M., Conan, G.Y., Robichaud, G., and Jones, A. 1991. Life history patterns and population fluctuations of snow crab (*Chionoecetes opilio*) in the fjord of Bonne Bay on the west coast of Newfoundland, Canada: from 1983 to 1990. Can. Tech. Rep. Fish. Aquat. Sci. No. 1817.

Conan, G.Y., and Comeau, M. 1986. Functional maturity and terminal molt of male snow crab, *Chionoecetes opilio*. Can. J. Fish. Aquat. Sci. **43**: 1710–1719.

Dickinson, J.L. 1986. Prolonged mating in the milkweed leaf beetle *Labdomera clivicollis clivicollis* (Coleoptera: Chrysomelidae): a test of the "sperm loading" hypothesis. Behav. Ecol. Sociobiol. **18**: 331–338.

Diesel, R. 1991. Sperm competition and the evolution of mating behavior in Brachyura, with special reference to spider crabs (Decapoda: Majidae). *In* Crustacean sexual biology. *Edited by* R.G. Bauer and J.W. Martin. Columbia University Press, New York. pp. 145–163.

Donaldson, W.E., and Adams, A.E. 1989. Ethogram of behavior with emphasis on mating for the tanner crab *Chionoecetes bairdi* Rathbun. J. Crustacean Biol. **9**: 37–53.

Drummond, B.A., III. 1984. Multiple mating and sperm competition in the Lepidoptera. *In* Sperm competition and the evolution of animal mating systems. *Edited by* R.L. Smith. Academic Press, New York. pp. 291–370.

Elner, R.W., and Beninger, P.G. 1992. The reproductive biology of snow crab, *Chionoecetes opilio*: a synthesis of recent contributions. Am. Zool. **32**: 524–533.

Elner, R.W., and Beninger, P.G. 1995. Multiple reproductive strategies in snow crab, *Chionoecetes opilio*: physiological pathways and behavioral plasticity. J. Exp. Mar. Biol. Ecol. **193**: 93–112.

Elner, R.W., Koshio, S., and Hurley, G.V. 1987. Mating behavior of the deep-sea red crab, *Geryon quinquedens* Smith (Decapoda, Brachyura, Geryonidae). Crustaceana (Leiden), **52**: 194–201.

Emlen, S.T., and Oring, L.W. 1977. Ecology, sexual selection, and the evolution of mating systems. Science (Washington, D.C.), **197**: 215–223.

Ennis, G.P., Hooper, R.G., and Taylor, D.M. 1988. Functional maturity in small male snow crabs (*Chionoecetes opilio*). Can. J. Fish. Aquat. Sci. **45**: 2106–2109.

Ennis, G.P., Hooper, R.G., and Taylor, D.M. 1990. Changes in the composition of snow crab (*Chionoecetes opilio*) participating in the annual breeding migration in Bonne Bay, Newfoundland. Can. J. Fish. Aquat. Sci. **47**: 2242–2249.

Gromko, M.H., Gilbert, D.G., and Richmond, R.C. 1984. Sperm transfer and use in multiple mating system of *Drosophila*. *In* Sperm competition and the evolution of animal mating systems. *Edited by* R.L. Smith. Academic Press, New York. pp. 371–426.

Halliday, T., and Arnold, S.J. 1987. Multiple matings by females: a perspective from quantitative genetics. Anim. Behav. **35**: 939–941.

Hartnoll, R.G. 1969. Mating in the Brachyura. Crustaceana (Leiden), **16**: 160–181.

Hinsch, G.W. 1968. Reproductive behavior in the spider crab *Libinia emarginata* (L.). Biol. Bull. (Woods Hole), **135**: 273–278.

Hooper, R.G. 1986. A spring breeding migration of the snow crab, *Chionoecetes opilio* (*O. fabr.*), into shallow water in Newfoundland. Crustaceana (Leiden), **50**: 257–264.

Hunter, F.M., Petrie, M., Otronen, M., Birkhead, T., and Møller, A.P. 1993. Why do females copulate repeatedly with one male? Trends Ecol. Evol. **8**: 21–26.

Jivoff, P. 1995. The role of mate guarding, male size and male investment on individual reproductive success in the blue crab, *Callinectes sapidus*. Ph.D. thesis, University of Maryland, College Park, Md.

Knudsen, J.W. 1964. Observations on the mating process of the spider crab *Pugettia producta* (Majidae, Crustacea). Bull. South. Calif. Acad. Sci. **63**: 38–41.

Kruse, G.H. 1993. Biological perspectives on crab management in Alaska. *In* Proceedings of the International Symposium on Management Strategies for Exploited Fish Populations. Alaska sea grant report No. 93-02. University of Alaska Sea Grant Program, Fairbanks, Alaska. pp. 355–384.

Laufer, H., and Ahl, J.S.B. 1995. Mating behavior and methyl farnesoate levels in male morphotypes of the spider crab, *Libinia emarginata* (Leach). J. Exp. Mar. Biol. Ecol. **193**: 15–20.

López-León, M.D., Cabrero, J., Pardo, M.C., Viseras, E., and Camacho, J.P.M. 1993. Paternity displacement in the grasshopper *Eyprepocnemis plorans*. Heredity, **71**: 539–545.

Lovrich, G.A., Sainte-Marie, B., and Smith, B.D. 1995. Depth distribution and seasonal movements of *Chionoecetes opilio* (Brachyura: Majidae) in Baie Sainte-Marguerite, Gulf of Saint Lawrence. Can. J. Zool. **73**: 1712–1726.

Moriyasu, M., and Conan, G.Y. 1988. Aquarium observation on mating behavior of snow crab, *Chionoecetes opilio*. ICES C.M. 1988/K:9. International Council for the Exploration of the Sea, Copenhagen, Denmark.

Orensanz, J.M., Parma, A.M., Armstrong, D.A., Armstrong, J., and Wardrup, P. 1995. The breeding ecology of *Cancer gracilis* (Crustacea: Decapoda: Cancridae) and the mating system of cancrid crabs. J. Zool. (London), **235**: 411–437.

Parker, G.A. 1970. Sperm competition and its evolutionary consequences in the insects. Biol. Rev. Camb. Philos. Soc. **45**: 525–567.

Paul, A.J. 1984. Mating frequency and viability of stored sperm in the Tanner crab *Chionoecetes bairdi* (Decapoda, Majidae). J. Crustacean Biol. **4**: 375–381.

Paul, A.J., Adams, A.E., Paul, J.M., and Feder, H.M. 1983. Some aspects of the reproductive biology of the crab *Chionoecetes bairdi*. Alaska sea grant report No. 83-1. University of Alaska Sea Grant Program, Fairbanks, Alaska.

Paul, A.J., Paul, J.M., and Donaldson, W.E. 1995. Shell condition and breeding success in Tanner crabs. J. Crustacean Biol. **15**: 476–480.

Sainte-Marie, B. 1993. Reproductive cycle and fecundity of primiparous and multiparous female snow crab, *Chionoecetes opilio*, in the northwest Gulf of Saint Lawrence. Can. J. Fish. Aquat. Sci. **50**: 2147–2156.

Sainte-Marie, B., and Carrière, C. 1995. Fertilization of the second clutch of snow crab, *Chionoecetes opilio*, mated once or twice after their molt to maturity. Fish. Bull. **93**: 758–763.

Sainte-Marie, B., and Hazel, F. 1992. Moulting and mating of snow crabs, *Chionoecetes opilio* (O. Fabricius), in shallow waters of the northwestern Gulf of Saint Lawrence. Can. J. Fish. Aquat. Sci. **49**: 1282–1293.

© 1997 NRC Canada

Sainte-Marie, B., and Lovrich, G.A. 1994. Delivery and storage of sperm at first mating of female *Chionoecetes opilio* (Brachyura, Majidae) in relation to size and morphometric maturity of male parent. J. Crustacean Biol. **14**: 508–521.

Sainte-Marie, B., Raymond, S., and Brêthes, J.-C. 1995. Growth and maturation of the benthic stages of male snow crab, *Chionoecetes opilio* (Brachyura: Majidae). Can. J. Fish. Aquat. Sci. **52**: 903–924.

Sainte-Marie, B., Sévigny, J.-M., Smith, B.D., and Lovrich, G.A. 1996. Recruitment variability in snow crab *Chionoecetes opilio*: pattern, possible causes, and implications for fishery management. *In* Proceedings of the International Symposium on the Biology, Management, and Economics of Crabs from High Latitude Habitats. Lowell Wakefield fisheries symposium series. Alaska sea grant report No. 96-02. University of Alaska Sea Grant Program, Fairbanks, Alaska. pp. 451–478.

Sévigny, J.-M., and Sainte-Marie, B. 1996. Electrophoretic data support the last-male sperm precedence hypothesis in the snow crab, *Chionoecetes opilio* (Brachyura: Majidae). J. Shellfish Res. **15**: 437–440.

Smith, R.L. 1980. Repeated copulation and sperm precedence: paternity assurance for a male brooding water bug. Science (Washington, D.C.), **205**: 1029–1031.

Stevens, B.G., Donaldson, W.E., Haaga, J.A., and Munk, J.E. 1993. Morphometry and maturity of paired tanner crabs, *Chionoecetes bairdi*, from shallow and deepwater environments. Can. J. Fish. Aquat. Sci. **50**: 1504–1516.

Takeshita, K., and Matsuura, S. 1980. Mating and egg-laying in Tanner crabs. Translation document No. 2314. International North Pacific Fisheries Commission, Vancouver, B.C.

Taylor, D.M., Hooper, R.G., and Ennis, G.P. 1985. Biological aspects of the spring breeding migration of snow crabs, *Chionoecetes opilio*, in Bonne Bay, Newfoundland (Canada). Fish. Bull. **83**: 707–711.

Thornhill, R., and Alcock, J. 1983. The evolution of insect mating systems. Harvard University Press, Cambridge, Mass.

Walker, W.F. 1980. Sperm utilization strategies in nonsocial insects. Am. Nat. **115**: 780–799.

Watson, J. 1970. Maturity, mating, and egg-laying in the spider crab, *Chionoecetes opilio*. J. Fish. Res. Board Can. **27**: 1607–1616.

Watson, J. 1972. Mating behavior in the spider crab, *Chionoecetes opilio*. J. Fish. Res. Board Can. **29**: 447–449.

Wilber, D.H. 1989. The influence of sexual selection and predation on the mating and postcopulatory guarding behavior of stone crabs (Xanthidae, Menippe). Behav. Ecol. Sociobiol. **24**: 445–451.

© 1997 NRC Canada

Clearance rates and filtering activity of zebra mussel (*Dreissena polymorpha*): implications for freshwater lakes

Martin J. Horgan and Edward L. Mills

Abstract: Zebra mussel (*Dreissena polymorpha*) filter-feeding impacts on phytoplankton in lakes depend on a number of factors including which phytoplankton are grazed and the population filtration rate, which in turn depends on individual clearance rate (volume of water cleared of particles per unit time) and the percentage of mussels filtering. We used short-term fixed-volume suspension depletion experiments to compare clearance of different seston particles and to concurrently measure clearance rates and the percentage of mussels filtering for different sizes of zebra mussel both day and night. Zebra mussel readily cleared particles ≤150 μm, including cyanobacterial filaments, and large mussels collected particles as large as 1.2 mm. Clearance rates did not differ among six differently shaped phytoplankton taxa. Filtering activity (percentage of mussels actively filtering) was 6–9% higher at night than during the day, but there were no diel changes in clearance rate. Clearance rate depended on mussel size, but filtering activity did not differ among shell length-classes. Filtering activity declined as zebra mussel depleted food concentration. Results suggest that filtering activity should be considered in population filtering impact assessments as a separate term from clearance rate because each may respond to different factors.

Résumé : L'impact de la moule zébrée (*Dreissena polymorpha*), qui s'alimente par filtration, sur le phytoplancton dans les lacs dépend d'un certain nombre de facteurs dont le type de phytoplancton brouté et le taux de filtration de la population qui dépend, à son tour, du taux d'élimination individuel (volume d'eau vidé de ses particules par unité de temps) et le pourcentage de moules qui filtrent. Nous avons utilisé des expériences d'épuisement des suspensions dans des volumes fixes à court terme pour comparer l'élimination de différentes particules de seston et pour mesurer, en même temps, les taux d'élimination et le pourcentage de moules qui filtrent pour différentes tailles de moules zébrées, le jour et la nuit. Les moules zébrées ont facilement éliminé des particules ≤150 μm, dont des filaments de cyanobactéries, et les moules de grande taille ont filtré des particules de taille aussi grosse que 1,2 mm. Les taux d'élimination n'ont pas différé pour six taxons de phytoplancton de morphologie différente. L'activité de filtration (pourcentage de moules faisant activement de la filtration) était plus élevée la nuit que le jour (augmentation de 6 à 9%), mais on n'observe pas de différences nycthémérales dans le taux d'élimination. Le taux d'élimination dépendait de la taille des moules, mais l'activité de filtration ne variait pas parmi les différentes classes de longueurs de coquilles. L'activité de filtration a diminué au fur et à mesure que les moules ont épuisé les aliments. Les résultats indiquent que l'activité de filtration devrait être prise en considération indépendamment du taux d'élimination dans les évaluations de l'impact de la filtration parce que ces deux éléments dépendent de facteurs différents.
[Traduit par la Rédaction]

Introduction

Recent colonizations of North American lakes by the exotic filter-feeding zebra mussel (*Dreissena polymorpha*) are likely to have significant effects on ecosystem processes (Nalepa and Schloesser 1993). Phytoplankton grazing by zebra mussel is of particular ecological concern because the mussels have high individual clearance rates (Kryger and Riisgård 1988) and reach high densities (e.g., $10^5/m^2$ in Lake Erie; Leach 1993). Zebra mussel are credited with causing recent reductions of phytoplankton biomass in Lake Erie of the Laurentian Great Lakes (MacIsaac et al. 1992; Leach 1993; Nicholls and Hopkins 1993; Holland 1993; Madenjian 1995; but see Wu and Culver 1991) and in an experimental pond in The Netherlands (Reeders et al. 1993). Information on the feeding biology of zebra mussel can help to assess their impact on phytoplankton in lakes.

To understand the effects of zebra mussel grazing on phytoplankton communities, it is important to determine which species they collect. Zebra mussel draw water through their mantle cavity, but use only a portion of the seston particles for their digestion while the rest are agglutinated as pseudofecal pellets and ejected (Stanczykowska and Planter 1985). Consequently, phytoplankton that is either digested or trapped in zebra mussel pseudofeces may constitute a significant loss of energy from the pelagia. Zebra mussel collect phytoplankton up to 750 μm (Ten Winkel and Davids 1982) and small zooplankton such as rotifers (MacIsaac et al. 1991) and then select particles of 15–40 μm for digestion once these are inside the mantle cavity (Ten Winkel and Davids 1982). Although much research has focused on clearance of small particles (e.g., Jørgensen et al. 1984; Sprung and Rose 1988), questions remain as to what sizes and shapes of larger phytoplankton,

Received October 31, 1995. Accepted July 15, 1996.
J13143

M.J. Horgan and E.L. Mills. Cornell Biological Field Station, Department of Natural Resources, Cornell University, 900 Shackelton Point Road, Bridgeport, NY 13030, U.S.A.

© 1997 NRC Canada

including cyanobacteria filaments, are cleared from the water by zebra mussel.

Estimates of size-specific zebra mussel clearance rates can be used to determine the extent of a mussel population's grazing impact on phytoplankton in lakes (e.g., MacIsaac et al. 1992). Zebra mussel clearance rates published in the literature have typically been derived from one of two types of studies. In the laboratory studies, clearance rates are measured using individual zebra mussel that are actively filtering (e.g., Kryger and Riisgård 1988; Sprung and Rose 1988; Bunt et al. 1993). Clearance rates are also reported from in situ studies in which groups of zebra mussel in containers are placed in a natural environment and the rate of depletion of natural seston is used to determine clearance rate (e.g., Stanczykowska et al. 1975; Reeders et al. 1989). Owing to the nature of the in situ studies, however, it is not possible to know whether all of the zebra mussel are actively filtering over the measurement period. It is known that zebra mussel punctuate their active filtering sessions with periods of quiescence during which their valves are closed and their siphons are withdrawn (Morton 1969). The percentage of time that zebra mussel are actively filtering is a complex variable behavior that has been related to time of day (Morton 1969; Walz 1978; Borcherding 1992), and possibly temperature, mussel size, and food concentration (Borcherding 1992). If the filtering activity in the in situ studies (such as Stanczykowska et al. 1975; Reeders et al. 1989) was less than 100%, the reported clearance rates are a composite of the percentage of mussels actively filtering and the rate at which actively filtering mussels pump water. For this reason, Morton (1971) adjusted his laboratory-derived clearance rates by half because he had previously found that his zebra mussel actively filtered only about 50% of the time (Morton 1969). In addition, Padilla et al. (1996) used a 67% adjustment to zebra mussel population filtration in their food web simulation model. Research that concurrently examines variation in clearance rate and filtering activity will provide information to help formulate accurate estimates of zebra mussel population filtering impact.

The objective of this study was to examine particle ingestion, filtering activity, and clearance rates for zebra mussel fed natural seston and artificial food. More specifically, we compared clearance rates of a wide size range of phytoplankton, from small unicells to filamentous taxa. We also measured the filtering activity and clearance rates for zebra mussel of different shell lengths during both day and night. In addition, we measured the response of filtering activity to a decline in food concentration. Both zebra mussel and natural seston used in experiments were from Oneida Lake, a large (20 700 ha), shallow, eutrophic lake in which zebra mussel were first found in 1991.

Methods

Acclimation of test animals

Zebra mussel were collected from rocks and native unionid clam shells from Oneida Lake, New York, 1992–1993. Individuals were detached by cutting the byssal threads with a razor blade and were placed in 4-L circular containers (14 cm deep, 19.5 cm in diameter). For all clearance rate experiments, zebra mussel were sorted into three size-classes: small (9–11 mm; 40/container), medium (13–15 mm; 20/container), and large (19–22 mm; 10/container). Smaller numbers of larger mussels were used to roughly compensate for their higher individual filtering rates. Mussels were fed natural seston from Oneida Lake for 5–7 days to acclimate feeding. Zebra mussel filter at lower rates than normal for at least 24 h after they are removed from the lake and taken to the laboratory (Reeders et al. 1989). During acclimation, water was exchanged in the experimental containers and fecal material was removed by siphoning once or twice a day. A 15 h light : 9 h dark cycle was maintained for acclimation and experimental periods. Temperature was maintained at 17 ± 1°C for all clearance rate experiments. Mussels were attached to the bottom of the containers and were consistently filtering after 5 days of acclimation.

Particle clearance experiments

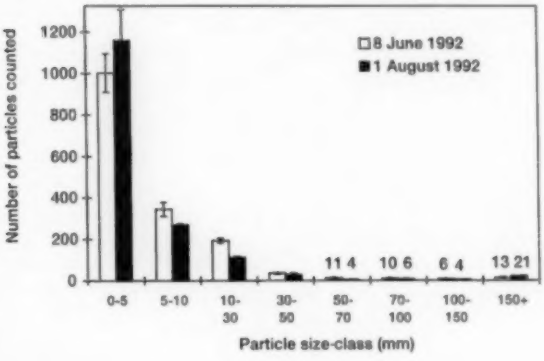
Zebra mussel clearance rates were calculated from the depletion of a number of phytoplankton taxa, particle size-classes, and cyanobacterial filaments from two assemblages of Oneida Lake natural seston. Differential selection of particles can be detected by comparing the depletion rate of different particle types from the same suspension. Measurements were made using clear-water (8 June 1992) and cyanobacterial (1 August 1992) assemblages. In June, clearance rates were determined for small- and medium-sized zebra mussel whereas in August, all three size-classes were included. Three replicate containers were used for each size-class in all experiments except in June when particle clearance rates were based on four replicates for small mussels. Initial and final seston samples (well-mixed 100-mL aliquots) were preserved in Lugol's solution for later analysis. Densities of three classes of natural seston particles in the samples were counted using an inverted microscope: (*i*) phytoplankton were identified to genus and species when possible and enumerated (Lund et al. 1958); (*ii*) 25 fields were examined at 40× and particles were size sorted (10–30, 30–50, 50–70, 70–100, 100–150, 150+ μm) by the greatest linear dimension (micrometres) for the small mussel size group (June only); and (*iii*) cyanobacterial filaments in 25 fields (August only) were counted.

The two phytoplankton assemblages fed to zebra mussel were chosen to differ in quantity and composition of phytoplankton. Phytoplankton counts were converted to biovolume using species-specific volumetric relationships (Wagner 1983). Phytoplankton biomass is expressed as fresh weight (micrograms) per litre by assuming 1.00 g/cm^3 algal volume. The phytoplankton assemblage in the June clear-water phase (total biomass = 1220 μg/L) was composed of primarily unicellular forms from four genera (61% of total biomass: *Cryptomonas*, *Mallomonas*, *Cyclotella*, and *Chlamydomonas*) and chain-forming diatoms (21% of total biomass: *Melosira* spp.). The August assemblage had a higher total biomass (4060 μg/L) and was dominated by two genera of filamentous cyanobacteria (58% of total biomass), *Aphanizomenon flos-aquae* and three species of *Anabaena*. The filamentous cyanobacteria appeared as individual filaments and were not clumped together. Colonial chlorophytes from the genera *Mougeotia*, *Coelastrum*, and *Eudorina* made up 20% of the total biomass in August, and cryptomonads accounted for an additional 9% of the biomass. Even though the species composition of the seston differed between the clear-water and cyanobacterial periods, particle size distributions between the two periods, as determined from microscopic counts, were similar (Fig. 1). Phytoplankton made up a small percentage of the total seston particle density (mean = 5.4% in June; mean = 5.9% in August), yet they likely made a greater contribution to the total seston biomass, since they were more represented in the large particle size-classes.

Direct observations were made of five byssal-attached mussels (10.1, 10.6, 13.4, 19.6, 20.5 mm shell length) fed colonies of *Gleotrichia echinulata* (0.4–1.2 mm in diameter). Each individual *G. echinulata* colony, measured with a Bausch and Lomb projecting microscope equipped with a stage micrometer, was presented to a single mussel for up to eight consecutive encounters. We recorded the behavioral response of the mussel to contact with each colony. A

© 1997 NRC Canada

Fig. 1. Mean numbers of seston particles (±1 SE) in eight size-classes counted in integrated water samples (0–11 m) collected from Oneida Lake on 8 June 1992 (open bars) and on 1 August 1992 (solid bars) and fed to zebra mussel in laboratory experiments. Numbers of particles for the four largest size-classes appear above the bars.



colony was considered collected if it passed through the incurrent siphon into the mantle cavity.

Clearance rates and filtering activity, day and night

We assessed clearance rate and filtering activity for zebra mussel day and night to determine whether there were diel changes in feeding behavior. Experiments were done once during the day (11:00) and once during the night (23:00) with the same individual mussels. Ground spinach was used as a test food because standard suspensions could be made for day and night experiments. A ground spinach suspension was made by homogenizing 10 g (wet weight) of fresh spinach leaves in 500 mL of filtered (Whatman 934-AH) Oneida Lake water for 2.5 min in a high-speed blender. The spinach concentrate was diluted with an additional 500 mL of water and passed through a 50-μm Nitex mesh to remove larger particles. Spinach concentrate (25 mL) was added to each 4-L experimental container at the beginning of the experiment. Three replicate containers were used for small, medium, and large zebra mussel shell length-classes. One-litre water samples from the beginning and end (after 2 h) of the experiments were analyzed for chlorophyll *a* to determine clearance rates (Strickland and Parsons 1972).

Filtering activity during food depletion

Filtering activity was observed as zebra mussel depleted a suspension of laboratory-cultured *Scenedesmus* spp. to see if filtering activity would change as food concentration was reduced. Two experimental containers with 25 zebra mussel (9–11 mm) were filled with a suspension of *Scenedesmus* spp. in 3 L of filtered Oneida Lake water. The filtering activity of zebra mussel and concentration of *Scenedesmus* spp. were determined over 34.0 h at intervals ranging from 0.3 to 10.0 h while temperature was 23 ± 3°C. *Scenedesmus* concentration was determined from 50-mL aliquots preserved in Lugol's solution; the concentration of algal cells in each sample was determined on a gridded microscope slide. The volume of water in each experimental container was kept constant by adding filtered Oneida Lake water.

Measuring filtering activity and clearance rate

We used the percentage of mussels with valves open and siphons extended as our measure of filtering activity. Over time, this measure should be equivalent to the percentage of time that the average mussel in the group is actively filtering. Filtering activity was observed in the dark using flashlights covered with red cellophane; zebra mussel were never observed to respond to the red light.

Clearance rate refers to the volume of water from which a mussel has removed all of the particles per unit time. Zebra mussel clearance rates were determined from the depletion of particles in a 4-L suspension for a 2-h period according to Coughlan (1969):

$$m = M \ln (\text{conc}_i/\text{conc}_f)(nt)^{-1}$$

where m is the individual clearance rate (volume per individual per hour), M is the volume of the suspension (millilitres), conc_i and conc_f are initial and final suspension concentrations, respectively, n is the number of individuals filtering, and t is time (hours). Suspension concentrations were either number of particles counted for particle clearance experiments or chlorophyll concentration (micrograms per litre) for day and night clearance rate experiments. The 2-h experimental period was chosen because it was long enough to measure significant particle depletion and short enough so that possible nutrient enrichment effects of zebra mussel on phytoplankton growth would be minimal. Control containers without zebra mussel in all experiments showed that adjustment for algal growth was not necessary. Each container with suspended seston was aerated and gently stirred every 0.5 h to prevent settling. Stirring the water caused only a slight disruption as a few mussels' valves closed but reopened in <60 s. For the experiments in which clearance rate and filtering activity were compared, the average individual clearance rate was calculated with the number of mussels actually filtering (n') by using the following formula:

$$n' = (\text{mussels/container}) \times \text{filtering activity (\%)}$$

Zebra mussel were observed every 0.5 h during these 2.0-h experiments and the average percentage of mussels filtering over four observations was used as the filtering activity for each experiment.

Data analysis

Clearance rates for zebra mussel on particles differing in size and type were compared using Kruskal–Wallis nonparametric analysis of variance (ANOVA) (Conover 1980). Day and night comparisons of filtering activity and clearance rate were made with repeated measures ANOVA (Neter et al. 1990) because experimental containers with zebra mussel were the same for both day and night. Curve fitting of the filtering activity data was done with a least-squares procedure (STATISTICA version 4.5).

Results

Clearance of natural seston

Results of particle clearance experiments indicate that zebra mussel (9–11 mm shell length) fed the June clear-water assemblage cleared particles ranging from 10 to over 150 μm at similar rates (Fig. 2). Mean clearance rates on particles in six size-classes were not significantly different (Kruskal–Wallis test; $p = 0.71$). Clearance rates were also compared for three shell length-classes of zebra mussel during June and August for six abundant phytoplankton taxa (Table 1). These taxa included *Chlamydomonas*, *Mallomonas*, *Cryptomonas pusilla*, *Cryptomonas ovata*, *Melosira*, and *Chroococcus*. There were no significant differences (Kruskal–Wallis; $p > 0.05$) among mean clearance rates for all six taxa examined (Table 1).

The clearance of colonies of *G. euchinulata* by zebra mussel was visually observed under a dissecting microscope. These particles were either taken into the mantle cavity

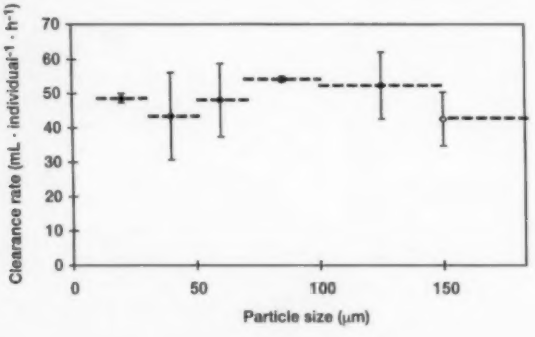
© 1997 NRC Canada

Table 1. Clearance rates (mL·individual^{-1}·h^{-1}) of six phytoplankton taxa and of cyanobacteria filaments.

	Zebra mussel shell length-class				
	Small (9–11 mm)		Medium (13–15 mm)		Large (19–22 mm)
	June	August	June	August	(August)
Chlorophyta					
Chlamydomonas sp. (small flagellate)	48±6	49±6	76±11	110±29	179±41
Chrysophyta					
Mallomonas sp. (large flagellate)	51±5		125±3		
Cryptophyta					
Cryptomonas pusilla (medium flagellate)	64±6	81±5	159±18	160±39	253±145
Cryptomonas ovata (large flagellate)	91±20	88±18		167±45	339±73
Bacillariophyta					
Melosira sp. (chainlike diatom)				114±37	
Cyanobacteria					
Chroococcus sp. (globular colony)		57±25			
Filaments		16±1		80±9	122±17
Kruskall–Wallis test (p value)	0.20	0.38	0.06	0.54	0.55

Note: Values are given as the mean ± 1SE. Clearance rates are presented for three shell length-classes of zebra mussel in June and August experiments. Kruskall–Wallis tests were used to compare clearance rates of different taxa (filaments not included).

Fig. 2. Mean clearance rates (±1 SE) of six size-classes of natural seston particles by small zebra mussel (9–11 mm). Solid symbols are at the midpoint of each size range; the open symbol is at the lower bound of the 150+ μm size-class. Broken horizontal lines indicate the range of particle sizes included in each size-class. Clearance rates were determined from experiments in which zebra mussel were fed a clearwater phytoplankton assemblage from Oneida Lake in June.



(collected) or rejected; the collection or rejection action sometimes occurred after the particle had remained at the outer edge of the incurrent siphon for several seconds. All five zebra mussel that were tested (10.1–20.6 mm) collected *G. euchinulata* colonies ≥0.9 mm in diameter; the largest colony collected was 1.2 mm, by a 20.6-mm zebra mussel. Most *G. euchinulata* colonies that were collected were subsequently ejected 30 s to a few minutes later.

Clearance rate and filtering activity, day and night

Repeated measures ANOVA of the experimental results revealed that clearance rates and filtering activity varied differently with mussel size and between day and night. Mean clearance rate was significantly different ($p > 0.01$) among size-classes, but did not differ significantly ($p = 0.48$) between day and night. However, filtering activity did not differ among mussel sizes ($p = 0.57$), but was significantly different between day and night ($p = 0.03$) (Table 2). Filtering activity was 6–9% higher during the night for all three size-classes of zebra mussel than during the day. Clearance rates for zebra mussel fed ground spinach were similar to those fed natural seston (Tables 1 and 2). Mean clearance rates for small, medium, and large zebra mussel fed ground spinach were 61, 110, and 217 mL·individual^{-1}·h^{-1}, respectively. These clearance rates are similar to the rates predicted from the allometric equation of Kryger and Riisgård (1988): 54, 110, and 247 mL·individual^{-1}·h^{-1} for the three average shell lengths.

Filtering activity during food depletion

A decline in filtering activity was observed for zebra mussel as they depleted a suspension of *Scenedesmus* spp. The data were

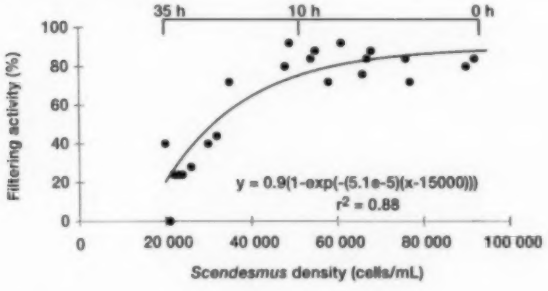
© 1997 NRC Canada

Table 2. Clearance rate (mL · individual^{-1} · h^{-1}) and filtering activity (%) for three shell length-classes of zebra mussel feeding on a suspension of ground spinach during both day and night.

	Zebra mussel shell length-class		
	Small (9–11 mm)	Medium (13–15 mm)	Large (19–22 mm)
Day			
Clearance rate	58±3	122±14	250±20
Filtering activity	72±3	75±4	79±9
Night			
Clearance rate	64±8	99±11	184±20
Filtering activity	80±2	84±1	85±5

Note: Values are given as the mean ± 1 SE.

Fig. 3. Filtering activity of zebra mussel plotted against food concentration as food is depleted through time (0–35 h) by filtering. Data are combined from two replicate experimental containers with 25 zebra mussel each.



fitted to an Ivlev's model with a correction for a nonzero x intercept (n = 22, r^2 = 0.88; Fig. 3). Filtering activity ranged from over 90% at high food concentrations to below 25% over 35 h of the experiment. *Scenedesmus* concentration was 9 × 10^5 cells/mL, initially, and was approximately 3 × 10^5 to 4 × 10^5 cells/mL when filtering activity declined sharply from about 80 to 40% (Fig. 3). Filtering activity was not observed after 35 h in this experiment, but observations of other zebra mussel that were not fed for up to 85 h suggest that mean filtering activity remains low and variable as low cell concentrations persist (M.J. Horgan, personal observation).

Discussion

Zebra mussel particle clearance

Our study results indicate that zebra mussel have similar clearance rates on a wide size range of natural seston particles as demonstrated by the clearance rates of 10- to 150-μm particles by 9- to 11-mm zebra mussel. We also observed 9- to 21-mm zebra mussel clearing 900 to 1200 μm diameter spherical particles; these particles are considerably larger than the previously reported largest particles ingested (750 μm, Ten Winkel and Davids 1982). In this study, phytoplankton morphology did not hamper clearance rates, as zebra mussel were able to ingest unicells, filaments, and globular colonies. In general, our results show that most particles of natural seston in freshwater lakes that are smaller than the incurrent siphon are subject to grazing by zebra mussel. These results are consistent with those of Nicholls and Hopkins (1993) who surveyed phytoplankton abundance trends and concluded that all of the dominant classes of algae in Lake Erie were equally affected by zebra mussel predation, including the large colony-forming diatoms.

Grazing by zebra mussel can have complex effects on the structure of natural phytoplankton communities. For example, phytoplankton that have adaptations that keep them away from the lake bottom such as swimming or buoyancy will be less susceptible to zebra mussel grazing (e.g., flagellated cryptophytes and cyanobacteria with gas vacuoles). Alternatively, phytoplankton such as diatoms that rely heavily on water mixing for water column positioning can settle to the lake bottom and suffer greater predation losses. Algal taxa that exhibit filament clumping such as *Aphanizomenon* can achieve a total particle size that is larger than the incurrent siphon of a zebra mussel. Blooms of clumped *Aphanizomenon* have occurred in Oneida Lake in late summer and early autumn since the establishment of zebra mussel. Such blooms of algae float to the lake surface and are both spatially unavailable and too large to be cleared by zebra mussel. Grazing of seston particles by zebra mussel adds a new predation pressure on the phytoplankton of many North American lakes. Our results indicate that zebra mussel are capable of collecting the majority of seston particles in freshwater lakes. Much of the collected seston will be rejected by the mussel within a pseudofecal matrix, and it is possible that some of the algal particles could remain viable and become resuspended into the water column. For example, the *G. echinulata* colonies collected by zebra mussel in this study were eventually ejected and appeared to be unaltered and viable.

Filtering activity and clearance rate

It has been reported that zebra mussel exhibit diel patterns in valve movements by actively filtering more during the night than during the day (Morton 1969). Borcherding (1992) also observed a similar diel pattern of filtering activity at low food concentrations (1.5 × 10^4 *Chlamydomonas* cells/mL), although he observed constant high filtering activity (80–90%) at higher food concentrations (2.5 × 10^4 cells/mL). Our result of 6–9% higher filtering activity at night than during the day supports the previous studies. The relatively small difference in filtering activity between day and night that we observed could be due to high food concentrations in our short-term experiments.

Filtering activity can also vary owing to food concentration. We observed a decline in filtering activity as food was depleted in experimental containers. A similar response was also observed in juvenile oysters, *Crassostrea virginica* (Higgins 1980). Our use of a functional response-type curve is not to suggest that we have measured a functional feeding response. The shape of the curve and the absolute values for filtering activity could be quite different if other initial concentrations of food were presented, or if food concentration was kept constant using a flow-through apparatus. The observed decline in filtering activity may have been due to satiation of the zebra mussel; however, the mussels always increased their filtering activity when food was added (M.J.

© 1997 NRC Canada

Horgan, personal observation). We believe that it is more likely that the reduction in filtering activity at low food concentrations was an energy-saving measure because there was low food reward for the energy expended in filtering. For suspension feeders such as zebra mussel, metabolic activity increases with clearance or feeding activity (Bayne et al. 1976). Even if large changes in filtering activity only occur at low food concentrations, this will be an important consideration because densely populated beds of suspension-feeding mussels are able to locally deplete concentrations of seston (Wildish and Kristmanson 1984; Frechette and Bourget 1985).

To our knowledge, no study has concurrently measured zebra mussel clearance rate and filtering activity and determined the contribution of both to overall filtering impact. Laboratory studies measuring clearance rate use actively filtering individuals and keep experimental conditions constant to eliminate variation owing to the environment. In situ studies that have reported variation in clearance rate (Stanczykowska et al. 1975; Reeders et al. 1989) typically report lower individual clearance rates than the laboratory studies (summarized in Kryger and Riisgård 1988). Our results showed that correcting a gross measure of clearance rate (comparable with results of in situ studies) by the filtering activity we were left with individual clearance rates similar to the high rates from previous laboratory studies. Because clearance rate varied with mussel size but not day and night, and filtering activity varied between day and night but not mussel size in our study, we conclude that these two feeding behaviors should be incorporated separately into estimates of the impact of zebra mussel population filtering.

Models of the impact of zebra mussel grazing can be made more accurate by taking variation in population filtering impact into account. Unfortunately, more research is needed to quantify the response of clearance rate and filtering activity to variable environmental conditions. There are a number of intrinsic and extrinsic factors that may cause variation in clearance rate and filtering activity for which evidence exists. The influence of zebra mussel size on clearance rate is well documented (see Kryger and Riisgård 1988) and our results suggest that mussel size has little effect on filtering activity within the range of sizes we tested. In contrast, Borcherding (1992) reported that 25-mm mussels had a lower filtering activity than 20-mm mussels; however, this conclusion was not statistically supported. Relationships between clearance rate and temperature have been reported (e.g., Morton 1971; Reeders and Bij de Vaate 1990) and filtering activity may be positively influenced by temperature (Borcherding 1992). Our results agree with those of studies that found a diel pattern in filtering activity (Morton 1969; Borcherding 1992), but we found no evidence that there is a diel pattern for clearance rate. Food concentration most likely plays an important role in both clearance rate (Walz 1978; Reeders et al. 1989; Sprung and Rose 1988) and filtering activity (Borcherding 1992; the present study). In addition, the type of food available to zebra mussel may influence clearance rate (Morton 1971; Stanczykowska et al. 1975), but whether it also affects filtering activity is uncertain.

Acknowledgments

This study was supported by funds from the New York Sea Grant Institute CR/CE-3 and is contribution No. 175 of the Cornell Biological Field Station. We thank Jacquelyn Rowe and Sandy Chin for their assistance with the laboratory experiments. We also thank Nelson Hairston Jr., Christine Mayer, and Lars Rudstam who reviewed earlier versions of the manuscript. The comments of Joseph Leach and two anonymous reviewers helped to improve the manuscript.

References

Bayne, B.L., Thompson, R.J., and Widdows, J. 1976. Physiology. I. *In* Marine mussels: their ecology and physiology. *Edited by* B.L. Bayne. IBP Handb. No. 10. Cambridge University Press, Cambridge. pp. 121–206.

Borcherding, J. 1992. Another early warning system for the detection of toxic discharges in the aquatic environment based on valve movements of the freshwater mussel *Dreissena polymorpha*. *In* The zebra mussel *Dreissena polymorpha*: ecology, biological monitoring and first applications in water quality management. *Edited by* D. Neumann and H.A. Jenner. Gustav Fischer Verlag, Stuttgart. pp. 127–146.

Bunt, C.M., MacIsaac, H.J., and Sprules, W.G. 1993. Pumping rates and projected filtering impacts of juvenile zebra mussel (*Dreissena polymorpha*) in western Lake Erie. Can. J. Fish. Aquat. Sci. **50**: 1017–1022.

Conover, W.J. 1980. Practical nonparametric statistics. 2nd ed. John Wiley & Sons, New York.

Coughlan, J. 1969. The estimation of filtering rate from the clearance of suspensions. Mar. Biol. (Berlin), **2**: 356–358.

Frechette, M., and Bourget, E. 1985. Energy flow between the pelagic and benthic zones: factors controlling particulate organic matter available to an intertidal mussel bed. Can. J. Fish. Aquat. Sci. **42**: 1158–1165.

Higgins, P.J. 1980. Effects of food availability on the valve movements and feeding behavior of juvenile *Crassostrea virginica* (Gmelin). II. Feeding rates and behavior. J. Exp. Mar. Biol. Ecol. **46**: 17–27.

Holland, R.E. 1993. Changes in planktonic diatoms and water transparency in Hatchery Bay, Bass Island Area, western Lake Erie since the establishment of the zebra mussel. J. Great Lakes Res. **19**: 617–624.

Jørgensen, C.B., Kiørboe, T., Møhlenberg, F., and Riisgård, H.U. 1984. Ciliary and mucus-net filter feeding, with special reference to fluid mechanical characteristics. Mar. Ecol. Prog. Ser. **15**: 283–292.

Kryger, J., and Riisgård, H.U. 1988. Filtration rate capacities in 6 species of European freshwater bivalves. Oecologia, **77**: 34–38.

Leach, J.H. 1993. Impacts of the zebra mussel, *Dreissena polymorpha*, on water quality and spawning reefs in western Lake Erie. *In* Zebra mussel: biology, impacts and control. *Edited by* T.F. Nalepa and D. Schloesser. Lewis Publishers, Boca Raton, Fla. pp. 381–397.

Lund, J.W.G., Kipling, C., and LeCren, E.D. 1958. The inverted microscope method of estimating algal numbers and the statistical basis of estimations by counting. Hydrobiologia, **11**: 142–170.

MacIsaac, H.J., Sprules, W.G., and Leach, J.H. 1991. Ingestion of small-bodied zooplankton by zebra mussel (*Dreissena polymorpha*): can cannibalism on larvae influence population dynamics? Can. J. Fish. Aquat. Sci. **48**: 2051–2060.

MacIsaac, H.J., Sprules, W.G., Johannsson, O.E., and Leach, J.H. 1992. Filtering impacts of larval and sessile zebra mussel (*Dreissena polymorpha*) in western Lake Erie. Oecologia, **92**: 30–39.

Madenjian, C.P. 1995. Removal of algae by the zebra mussel (*Dreissena polymorpha*) population in western Lake Erie: a bioenergetics approach. Can. J. Fish. Aquat. Sci. **52**: 381–390.

© 1997 NRC Canada

Morton, B. 1969. Studies on the biology of *Dreissena polymorpha* Pall. II. Correlation of the rhythms of adductor activity, feeding, digestion and excretion. Proc. Malacol. Soc. Lond. **38**: 401–414.

Morton, B. 1971. Studies on the biology of *Dreissena polymorpha* Pall. V. Some aspects of filter-feeding and the effect of micro-organisms upon the rate of filtration. Proc. Malacol. Soc. Lond. **39**: 289–300.

Nalepa, T.F., and Schloesser, D.W. (*Editors*). 1993. Zebra mussel: biology, impacts, and control. Lewis Publishers, Boca Raton, Fla.

Neter, J., Wasserman, W., and Kutner, M.H. 1990. Applied linear statistical models. 3rd ed. Irwin, Homewood, Ill.

Nicholls, K.H., and Hopkins, G.J. 1993. Recent changes in Lake Erie (north shore) phytoplankton: cumulative impacts of phosphorus loading reductions and the zebra mussel introduction. J. Great Lakes Res. **19**: 637–647.

Padilla, D.K., Adolph, S.C., Cottingham, K.L., and Schneider, D.W. 1996. Predicting the consequences of dreissenid mussels on a pelagic food web. Ecol. Model. **85**: 129–144.

Reeders, H.H., and Bij de Vaate, A. 1990. Zebra mussel (*Dreissena polymorpha*): a new perspective for water quality management. Hydrobiologia, **200–201**: 437–450.

Reeders, H.H., Bij de Vaate, A., and Slim, F.J. 1989. The filtration rate of *Dreissena polymorpha* (Bivalvia) in three Dutch lakes with reference to biological water quality management. Freshwater Biol. **22**: 133–141.

Reeders, H.H., Bij de Vaate, A., and Noordhuis, R. 1993. Potential of the zebra mussel (*Dreissena polymorpha*) for water quality management. *In* Zebra mussel: biology, impacts, and control. *Edited by* T.F. Nalepa and D.W. Schloesser. Lewis Publishers, Boca Raton, Fla. pp. 439–451.

Sprung, M., and Rose, U. 1988. Influence of food size and food quantity on the feeding of the mussel *Dreissena polymorpha*. Oecologia, **77**: 526–532.

Stanczykowska, A., and Planter, M. 1985. Factors affecting nutrient budget in lakes of the R. Jorka watershed (Masurian Lakeland, Poland). X. Role of the mussel *Dreissena polymorpha* (Pall.) in N and P cycles in a lake ecosystem. Ekol. Pol. **33**: 345–356.

Stanczykowska, A., Lawacz, W., and Mattice, J. 1975. Use of field measurements of consumption and assimilation in evaluation of the role of *Dreissena polymorpha* Pall. in a lake ecosystem. Pol. Arch. Hydrobiol. **22**: 509–520.

Strickland, J.D.H., and Parsons, T.R. 1972. A practical handbook of seawater analysis. 2nd ed. Bull. Fish. Res. Board Can. No. 167.

Ten Winkel, E.H., and Davids, C. 1982. Food selection by *Dreissena polymorpha* Pallas (Mollusca: Bivalvia). Freshwater Biol. **12**: 553–558.

Wagner, K.J. 1983. The impact of natural phytoplankton assemblages on *Daphnia pulex* reproduction in Oneida Lake, New York. M.S. thesis, Cornell University, Ithaca, N.Y.

Walz, N. 1978. The energy balance of the freshwater mussel *Dreissena polymorpha* Pallas in laboratory experiments and in Lake Constance. I. Pattern of activity, feeding and assimilation efficiency. Arch. Hydrobiol. Supplbd. **55**: 83–105.

Wildish, D.J., and Kristmanson, D.D. 1984. Importance to mussels of the benthic boundary layer. Can. J. Fish. Aquat. Sci. **41**: 1618–1625.

Wu, L., and Culver, D.A. 1991. Zooplankton grazing and phytoplankton abundance: an assessment before and after invasion of *Dreissena polymorpha*. J. Great Lakes Res. **17**: 425–436.

© 1997 NRC Canada

Reproductive characteristics of the ruffe, *Gymnocephalus cernuus*, in the St. Louis River estuary on western Lake Superior: a histological examination of the ovaries over one annual cycle

Richard L. Leino and J. Howard McCormick

Abstract: The Eurasian ruffe, *Gymnocephalus cernuus*, is a small percid that has recently been introduced into the Duluth-Superior Harbor at the western end of Lake Superior. Because of its adverse impact on native species, its presence has caused considerable concern in the North American fisheries community. Ruffe have experienced phenomenal reproductive success in their new home and are rapidly extending their range. The reproductive success of this species seems to be partly related to the manner of spawning. We describe the dynamics of oogenesis of different age groups during the 1994 reproductive cycle. Spawning commenced within a week or two of ice-out and extended for approximately 8 weeks. Histological observations revealed that different age groups of ruffe spawned somewhat in succession, with older fish spawning first. As the season progressed, earlier spawners engaged in repeat spawning, probably two or three times before the end of the spawning period. These observations suggest that the unstable spring weather characteristic of this region has a minimal effect on the success of ruffe age groups, in contrast with several native species that spawn only once in the spring and that could suffer losses of age groups during a postspawning cold spell.

Résumé : La grémille eurasienne (*Gymnocephalus cernuus*) est un petit percidé qui a récemment été introduite dans le port de Duluth-Superior à l'extrémité occidentale du lac Supérieur. À cause de ses effets défavorables sur les espèces indigènes, cette espèce a suscité de fortes préoccupations auprès de la communauté halieutique de l'Amérique du Nord. La grémille s'est multipliée de manière phénoménale dans son nouveau milieu et est en train d'étendre rapidement son aire de distribution. Le succès de cette espèce du point de vue de la reproduction semble partiellement lié à son mode de frai. Nous avons décrit la dynamique de l'oogenèse chez différents groupes d'âges durant le cycle de reproduction de 1994. Le frai a débuté 1 ou 2 semaines après le déglacement et s'est poursuivi pendant environ 8 semaines. L'examen histologique a révélé que différents groupes d'âges de grémille ont frayé quelque peu successivement, avec les poissons plus agés frayant les premiers. Comme la saison s'avançait, les frayeurs primeurs frayaient plusieurs fois, probablement deux ou trois fois, avant la fin du frai. Ces constatations donnent à penser que l'instabilité du climat dans cette région au printemps n'a qu'un effet négligeable sur le succès reproductif des groupes d'âges de la grémille, contrairement à ce qui se passe pour plusieurs espèces indigènes qui ne frayent qu'une seule fois au printemps et qui pourraient subir la perte de groupes d'âges au cours d'une période de froid suivant le frai. [Traduit par la Rédaction]

Introduction

Ruffe, *Gymnocephalus cernuus*, were introduced into North America in or shortly prior to 1986 when young of the year were first collected in the Duluth-Superior Harbor (DSH), a part of the St. Louis River estuary at the western end of Lake Superior (Pratt et al. 1992). This harbor is an active inland seaport receiving ships from throughout the world. Since 1986, ruffe have appeared elsewhere in Lake Superior and are becoming established along its south shore and in Thunder Bay, Ont. Recently, they have been collected in Lake Huron near Alpena, Mich. (J.H. Selgeby, Ashland Biological Station, Ashland, Wis., personal communication). One reason that this range expansion is of major concern is that ruffe are believed to have great potential for damaging indigenous fisheries, especially for percids in the lower Great Lakes and in inland lakes and rivers (Ogle 1995). For example, in the DSH, as the ruffe population expanded, yellow perch, *Perca flavescens*, trout-perch, *Percopsis omiscomaycus*, and several cyprinid populations declined severely (Selgeby 1995). Moreover, ruffe have not been well utilized as a replacement forage by walleye, *Stizostedion vitreum*, and other game fish (Ogle et al. 1996) and are unattractive to fisherman because of their small size.

The reproductive cycle of ruffe in its new North American home is of considerable interest, particularly with respect to the timing of spawning and spawning strategy. For example, knowledge regarding ruffe spawning habits would be of value

Received April 9, 1996. Accepted July 15, 1996.
J13394

R.L. Leino. Department of Anatomy and Cell Biology, School of Medicine, University of Minnesota, Duluth, MN 55812, U.S.A.
J.H. McCormick. U.S. Environmental Protection Agency, Mid-Continent Ecology Division, Duluth, MN 55804, U.S.A.

© 1997 NRC Canada

in strategic planning involving applications of piscicides. Because ruffe eggs are more resistant to these chemicals than are the fish, it is critical to know when eggs are present in the environment (Fairchild and McCormick 1996).

In Europe, ruffe reportedly are capable of spawning twice in a given year. Koshelev (1963) provided evidence that in 1958 in Lake Glubokoe, north of Moscow, ruffe spawned once from mid- to late May and again in late June and July. In 1959, they also spawned twice, but began a week earlier in both cases. Survey data (Selgeby 1995) suggest that ruffe may spawn only once in the DSH. However, no direct observations of ruffe spawning have been reported. The survey data consist of gonadosomatic indices (GSIs), which may or may not be reliable indicators of spawning (deVlaming et al. 1982). For example, ruffe may be capable of releasing eggs from moderately sized ovaries. A more certain method of determining spawning capability in ruffe is by histological examination of their ovaries (Koshelev 1963), the objective of the present study. We also compare these histologically staged ovaries with the respective GSIs of the same fish to determine if the corresponding GSI determinations would provide similar assessments of spawning states.

Materials and methods

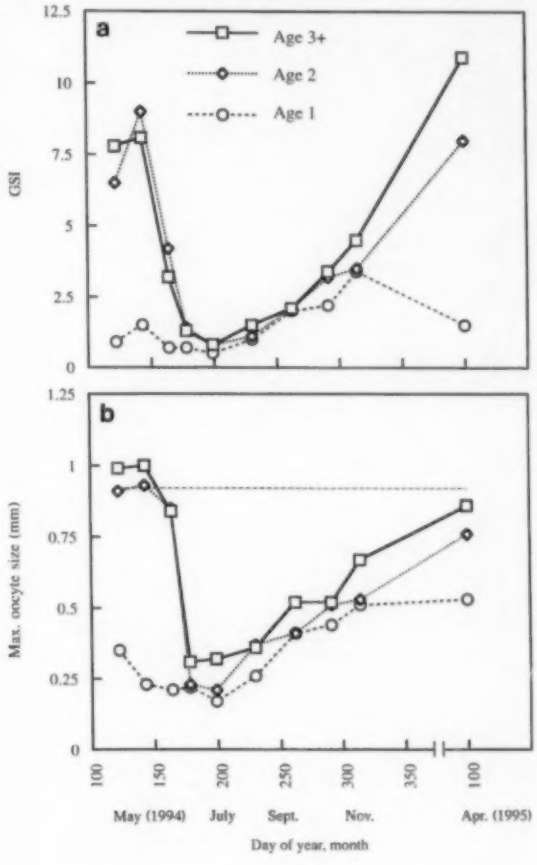
Ruffe used for ovarian examinations were collected at nine regularly spaced intervals from early May to near ice-up in mid-November 1994 and once at ice-out in April 1995 (Table 1). These collections were made with bottom trawls at two general locations in the DSH, one in the vicinity of the Blatnik Bridge and the other near the ore docks in Allouez Bay. Individuals from three size groups of female ruffe (small, medium, and large) were collected, weighed, and measured (total length, TL) during each sampling period to insure an adequate representation of fish from different age groups. The ruffe from these size groups were aged via scale analysis and divided into age groups: ruffe in their 4th year of life or older (age 3+; approximately 75% of these similar-sized fish were age 3 and 25% were age 4) and those in their 3rd (age 2), 2nd (age 1), and 1st (age 0) years (Table 1). Ovaries were preserved in 2.5% glutaraldehyde – 2% formaldehyde in 0.08 M phosphate buffer, pH 7.4 (Leino et al. 1990), processed into JB-4 methacrylate embedding media, sectioned at 3 μm, and stained with hematoxylin and eosin. Little oocyte shrinkage occurs with this method.

Four cross sections of ovaries from each fish were microscopically examined and staged using criteria based on studies of Selman and Wallace (1986) on mummichog, *Fundulus heteroclitus*, and Treasurer and Holliday (1981) on river perch, *Perca fluviatilis* (Table 2). The same sections were used to measure the diameters of oocytes in various stages of development. For the data on the diameter of the largest oocytes observed (Table 1), the largest oocyte, of several similar-sized oocytes measured with an eyepiece micrometer, was selected for each fish. (The largest oocytes were sectioned very near their centers, e.g., compared with more peripherally sectioned oocytes, they had thin, sharply defined vitelline envelopes and large, sharply defined germinal vesicles.) Some ovaries were also sectioned longitudinally to confirm that oocyte distribution was similar throughout the length of the ovary.

Results

In 1994, ruffe spawned from late April (some ovaries from the May 2 sampling period had corpora lutea (postovulatory follicles)) through mid-June. There was no evidence that ruffe have a July spawning period in the DSH (Fig. 1), but the single spawning period is lengthy, at least 8 weeks, and some ruffe appeared to release more than one batch of eggs during that time. The following summary details the seasonal ovarian changes of DSH ruffe for different age groups during the 1994 reproductive season and into the following spring (Table 1).

Fig. 1. (*a*) Mean GSI (= mass of ovary/mass of fish × 100) of ruffe used in the present study. (*b*) Mean diameters of the largest oocyte found in the ovarian sections for each ruffe examined. The broken horizontal line denotes a spawning-size oocyte.



May 2

The surface water temperature was 7.4°C; most spawning occurred at <1 m in the DSH (Selgeby 1995). Spawning in age 3+ fish was well underway by May 2 on the basis of the presence of stage 5 oocytes and (or) corpora lutea (Figs. 2, 3*a*, 4*b*) in 5 of 10 fish. The ovaries contained oocytes in several stages of development (Figs. 2, 3*b*), a characteristic of serial or batch spawners. Oocyte maturity of age 2 fish was slightly less advanced than that of age 3+ fish and no ovaries had corpora lutea. All age 1 females had immature stage 1 or 2 (as in Fig. 3*c*) ovaries. Some of this age group's ovaries, which had advanced to stage 3 over winter, exhibited atresia of the stage 3 oocytes.

© 1997 NRC Canada

Table 1. Total numbers and mean mass (SD in parentheses) of ruffe examined, and mean diameters (range in parentheses) of the largest oocyte observed in the ovarian sections for each ruffe compared with stages present on dates sampled.

Date	Age group	*N*	Mean mass (g)	Oocyte size (μm)	Stages
May 2, 1994	3+	9	22.8 (6.1)	807 (442–990)	1–5
	2	11	7.4 (2.2)	645 (20–913)	1–4A
	1	10	2.1 (0.4)	168 (13–350)	1–2
May 23, 1994	3+	10	19.9 (4.9)	802 (673–1005)	1–5
	2	10	9.3 (2.1)	771 (650–930)	1–5
	1	10	2.5 (0.8)	140 (105–230)	1–2
June 13, 1994	3+	10	18.7 (4.4)	524 (200–836)	1–4A
	2	11	7.6 (1.9)	584 (65–845)	1–4A
	1	9	3.6 (0.5)	147 (105–206)	1–2
June 27, 1994	3+	11	26.0 (9.1)	234 (140–310)	1–2
	2	7	8.6 (1.1)	193 (160–234)	1–2
	1	9	4.6 (0.8)	149 (115–215)	1–2
July 18, 1994	3+	11	26.0 (5.2)	202 (180–320)	1–2
	2	8	9.4 (3.3)	172 (150–210)	1–2
	1	6	5.4 (1.3)	152 (130–173)	1–2
Aug. 18, 1994	3+	9	24.3 (5.0)	291 (205–357)	1–2
	2	9	12.4 (4.4)	258 (163–373)	1–2
	1	6	6.1 (1.4)	222 (195–259)	1–2
Sept. 19, 1994	3+	11	22.0 (5.5)	400 (314–515)	1–3E
	2	8	14.2 (4.7)	347 (290–392)	1–2
	1	9	7.9 (1.7)	328 (250–407)	1–2
	0	6	4.7 (0.7)	202 (105–280)	1–2
Oct. 18, 1994	3+	6	21.9 (4.1)	452 (410–516)	1–2
	2	5	17.1 (5.5)	445 (382–505)	1–2
	1	5	7.0 (1.7)	367 (254–440)	1–2
	0	5	5.2 (1.2)	398 (352–428)	1–2
Nov. 10, 1994	3+	8	21.8 (5.5)	528 (410–674)	1–3E
	2	8	10.5 (3.3)	459 (355–530)	1–3E
	1	7	10.2 (1.9)	453 (402–529)	1–3E
	0	8	4.9 (0.9)	396 (290–4609)	1–2
Apr. 10, 1995	3+	10	33.0 (5.2)	815 (731–860)	1–3L
	2	9	9.2 (1.9)	692 (565–755)	1–3L
	1	10	2.9 (0.8)	252 (120–534)	1–3E

Note: E, early in stage; L, late in stage.

May 23

The water temperature was 17.0°C. Age 3+ and some age 2 fish were spawning, and several oocyte stages were present in a typical ovary (Fig. 3*d*). All age 1 fish had immature stage 1 or 2 ovaries.

June 13

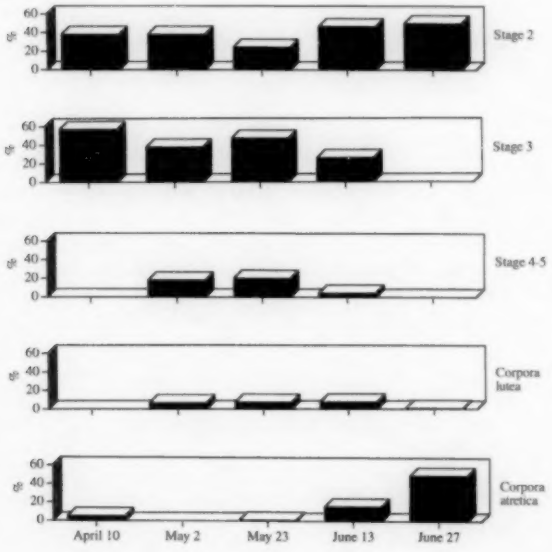
The water temperature was 19.0°C. Eight of 10 age 3+ ruffe appeared to have recently completed their spawning. Their ovaries contained corpora lutea and unspawned oocytes. These oocytes were becoming atretic rather than progressing to maturity (Figs. 2, 3*e*, 4*c*). The other two fish were nearly ready to spawn. Two of 11 age 2 fish had spawned. Five age 2 fish were in the process of resorbing large numbers of unspawned oocytes, i.e., they had numerous preovulatory atretic follicles (corpora atretica) (Fig. 3*f*). These fish lacked corpora lutea, suggesting that they did not spawn. Four other age 2 fish had nearly mature late stage 3 ovaries and had not

© 1997 NRC Canada

Table 2. Histological stages used for classification of ruffe ovarian development (primarily based on Selman and Wallace 1986).

Stage	Characteristics
1. Primary growth	Oogonia, small primary oocytes
1A	Oocytes in nests; nuclei in prophase I pattern
1B	Oocytes larger, out of nests, surrounded by follicle cells; many pleiomorphic nucleoli
2. Cortical alveolus	Appearance of cortical alveoli and small lipid droplets (Fig. 3*c*)
3. Vitellogenic	Appearance of yolk bodies (Fig. 3*i*)
4. Oocyte maturation	Nucleus (germinal vesicle) disappears, indicating resumption of meiosis
4A	Yolk bodies not fused (Fig. 3*d*)
4B	Yolk bodies substantially fused; large lipid droplet formed (Fig. 3*b*)
5. Mature egg – spawning	Oocyte appearance similar to 4B; intense vasodilation in ovary; oviduct wall very thin

Fig. 2. Oocyte stages present just prior to, during, and just after spawning in age 3+ ruffe. Note that preovulatory atretic follicles were present in the April sample, perhaps because many of these fish were nearly ready to spawn, but, presumably, the water temperature (1°C) or other variables were not suitable. At the end of spawning, a massive atresia of the remaining late stage 2, stage 3, and stage 4+ oocytes occurred.



yet spawned. All age 1 fish had immature stage 1 or 2 (Fig. 3*c*) ovaries. Oocytes that had advanced to a certain size in stage 2 appeared to undergo atresia rather than to enter stage 3. We found no evidence at any time that age 1 fish had spawned.

June 27

The water temperature was 23.0°C. Spawning of age 3+ ruffe appeared to be completed. All ovaries had large numbers of unspawned late stage 2 and older oocytes that were undergoing atresia (as in Fig. 3*e*). Possible corpora lutea were recognized in only 1 of the 10 ovarian samples. (Old corpora lutea may have been present, but they were difficult to distinguish from old preovulatory atretic follicles.) Many of the next year's group of oocytes were in early to middle stage 2. Most age 2 fish were resorbing large numbers of stage 3 oocytes. Few ovaries of age 2 fish appeared to have advanced beyond stage 3. Numerous healthy next year's stage 2 oocytes were present among the atretic stage 3 oocytes. Age 1 fish had ovaries with small stage 2 oocytes.

July 18

The water temperature was 20.5°C. All age groups had ovaries with stage 2 oocytes that were larger than on June 27. The mean size of oocytes of age 3 fish was greater than that of age 2 fish, whose oocytes were larger than those of age 1 fish (Table 1). Atresia of older oocytes left over from spawning was nearly completed.

August 18

The water temperature was 19.5°C. All age groups had ovaries with growing stage 2 oocytes. The oocyte size differential between age groups was maintained.

September 19

The water temperature was 17.0°C. All age groups showed further growth of stage 2 oocytes, maintaining the size differential between age groups. Young-of-the-year (age 0) ruffe sampled on this date also had stage 2 oocytes, although they were much smaller than those of older age groups.

© 1997 NRC Canada

Fig. 3. (*a*) Corpora lutea (postovulatory follicle, arrowhead) in the ovary of an age 3 ruffe, May 2 sample, that had recently spawned. Also present are late stage 3 oocytes (arrow) representing the next wave of ovulation in this serial spawner. Scale bar = 74 μm. (*b*) Oocytes in several stages of development are present in this stage 4B ovary from an age 3 fish, May 2 sample. Scale bar = 370 μm. (*c*) Ovary of an age 1 ruffe, June 13 sample. Ovaries of age 1 fish did not advance beyond stage 2 during the spawning period. Scale bar = 147 μm. (*d*) Ovary of an age 3 ruffe, May 23 sample, that had spawned previously (corpora lutea, not shown, were present) showing another clutch of oocytes (stage 4A, arrowhead) nearing maturity. Scale bar = 147 μm. (*e*) Ovary of an age 3 ruffe, June 13 sample, showing atresia of unspawned oocytes (arrowheads) in this postspawning ovary. Scale bar = 137 μm. (*f*) Ovary of an age 2 ruffe, June 13 sample, showing atresia of the closely packed unspawned oocytes. There was no evidence that this fish had spawned (there were no corpora lutea found). Scale bar = 338 μm. (*g*) Ovary of an age 3 ruffe, November 10 sample. Many of the oocytes have reached early stage 3 (arrowhead). Scale bar = 147 μm. (*h*) Ovary of an age 2 ruffe, November 10 sample. This stage 2 ovary is behind that of age 3+ fish in development. Scale bar = 147 μm. (*i*) Ovary of an age 3 ruffe, April 10 sample. Many oocytes have reached late stage 3. Scale bar = 147 μm. (*j*) Ovary of an age 2 ruffe, April 10 sample. Oocytes are smaller and less mature than in age 3+ fish in this early to middle stage 3 ovary. Scale bar = 147 μm.

October 18

The water temperature was 14.5°C. All groups showed further growth of stage 2 oocytes. Development of oocytes of age 2 fish had caught up to that of oocytes of age 3 fish. Oocytes of age 0 fish had caught up to those of age 1 ruffe, whose oocytes remained smaller than those of older year-classes.

November 10

The water temperature was 6.0°C. Most age 3+ ruffe had early stage 3 ovaries (Fig. 3*g*). Most age 2 fish had ovaries that were still in stage 2 (Fig. 3*h*). The mean diameter of the largest oocytes of individual fish was 87% of that of age 3+ fish. That is, oocytes of age 2 fish again fell behind those of age 3+ fish in development. On the other hand, ovarian development of age 1 fish had caught up to that of age 2 fish. Ovarian development of age 0 ruffe was not far behind that of age 1 fish.

April 10

The water temperature was 1.0°C. Oocytes of age 3+ ruffe continued to grow over the winter and were in late stage 3, prespawning size (Figs. 3*i*, 4*a*). Oocytes of age 2 fish also grew during the winter, but their mean diameter was only 85% of that of age 3+ fish (Fig. 3*j*; Table 1). Most age 1 ruffe had stage 2 ovaries, but lagged far behind older fish in oocyte size.

Discussion

Histological examination of ruffe ovaries over an annual cycle strongly suggests that, as in many parts of Europe (Kovalev 1973; Craig 1987; Neja 1988; Ogle 1995), ruffe are batch spawners in the DSH. Their ovaries may contain several oocyte stages simultaneously. The oocytes occur in group-synchronous, multiple clutches, a pattern also observed, for example, in white perch, *Morone americana* (Jackson and Sullivan 1995). This pattern is particularly evident in larger ruffe, which appear to release more than one clutch of eggs during the long spawning period. This spawning strategy gives greater assurance of consistent annual reproductive success. It provides a second chance, and probably even multiple chances, for eggs produced in a given year to experience suitable incubation conditions for successful hatching. This is of particular importance in areas with short growing seasons and with highly variable spring weather (Kramer and Smith 1962; Koonce et al. 1977).

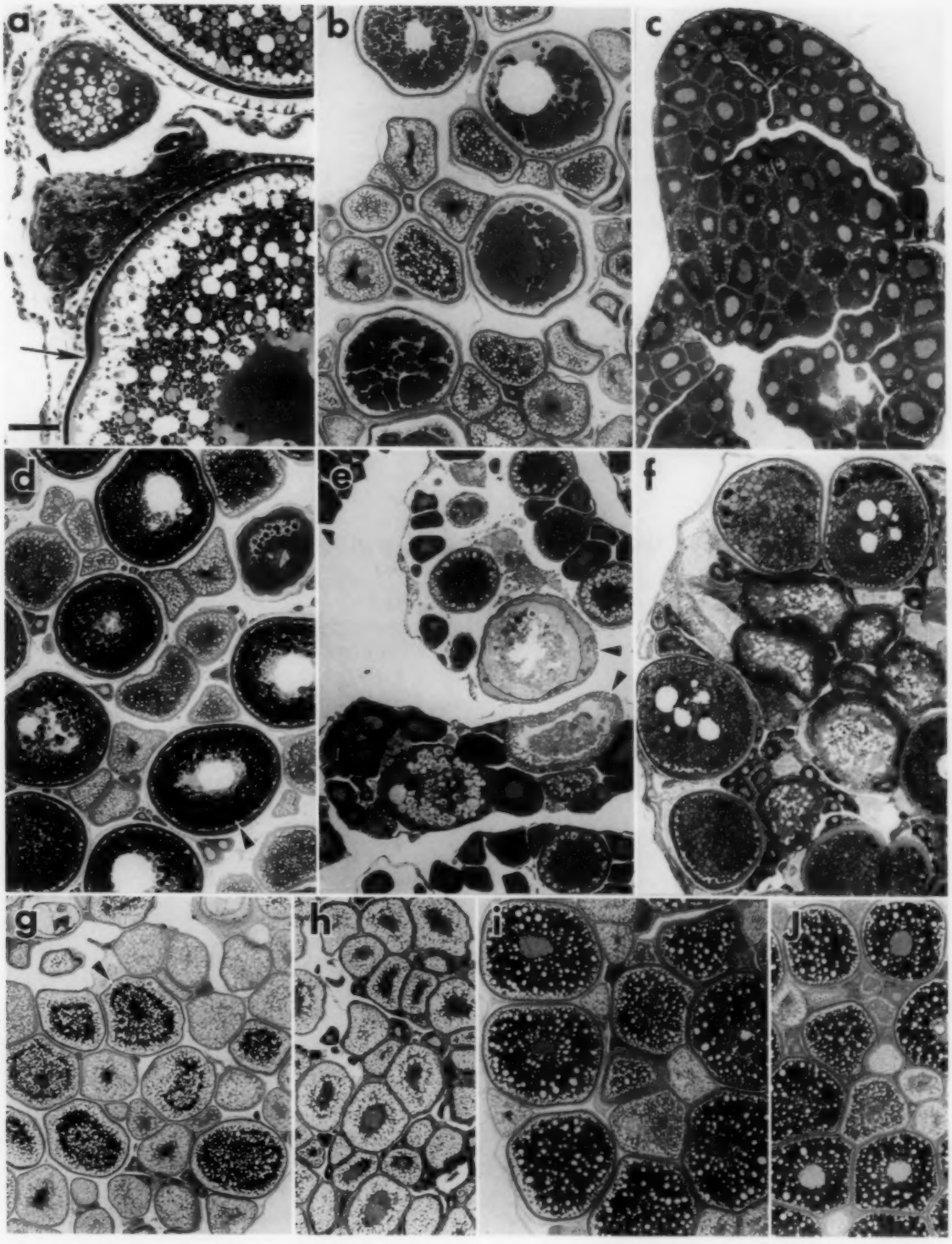
The large number of nearly mature oocytes left over after spawning may suggest that ruffe are poorly adapted to their new environment. Production of unspawned oocytes is energetically expensive, although part of this energy can be recovered through oocyte resorption. On the other hand, overproduction of oocytes in batch spawners could occur so that they are prepared to spawn whenever and for as long as environmental conditions are appropriate. Ruffe in parts of their native habitat, presumably where they are well adapted, also exhibit this postspawning oocyte overproduction (V. Kovac, Comenius University, Bratislava, Slovakia, personal communication).

Histological examination of ruffe from the DSH population revealed that the spawning period extended from late April through mid-June in 1994. There was no evidence of a second spawning period, e.g., from late June into July, as reported for ruffe in some Russian reservoirs (Koshelev 1963). Instead, maturing eggs present in the ovaries either were spawned in one or more batches by mid-June or underwent atresia and were resorbed. By mid-June, water temperatures exceeded normal spawning temperatures (Ogle 1995) for ruffe, and this could account for the ovarian regression.

Most spawning in the DSH involved age 3+ and older females. (Age 1 and older male ruffe had plentiful sperm in their testes throughout the spawning season (R.L. Leino, unpublished data).) Few, perhaps only 20%, of age 2 females appeared to have spawned. Their ovarian development lagged behind that of older ruffe in the spring. This resulted in ovulation in only some of these fish and then only near the end of the spawning period. Consequently, most age 2 females that did spawn appeared to have released only a single clutch of eggs. In most age 2 ruffe, stage 3 and 4 oocytes underwent atresia rather than progressing to ovulation. Age 1 ruffe did not appear to contribute to spawning in the DSH. Their oocytes did not advance beyond stage 2 during the spawning period. There was extensive resorption of these stage 2 oocytes near the end of the spawning period.

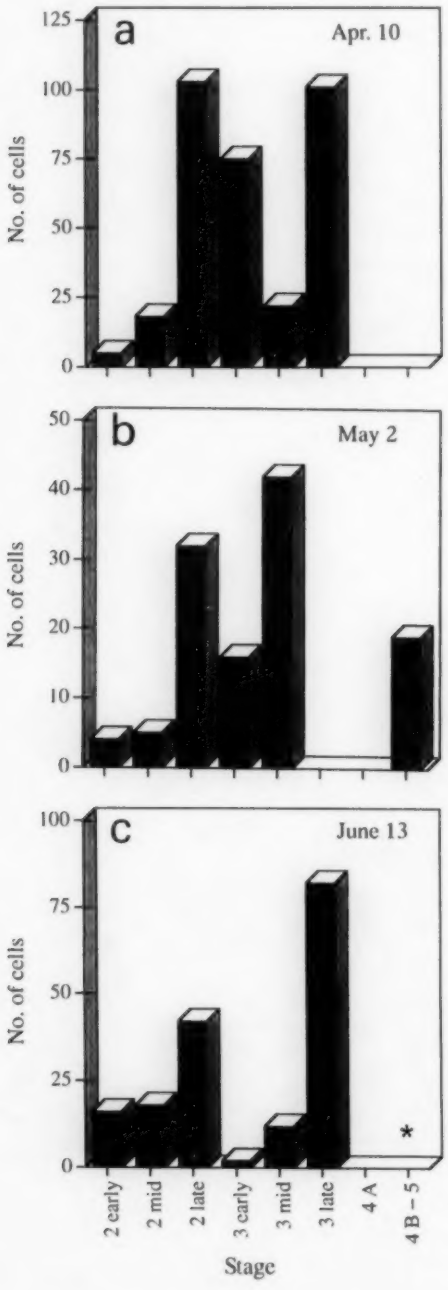
In the present study the GSI, while usually adequate for assessing the spawning state of the ruffe population, was not a dependable indicator of spawning readiness of individual fish or for some age groups. For example, in four ruffe that were histologically on the verge of spawning (stage 4B or 5) the GSI ranged from 6.3 to 21.7, while the GSI of stage 3 ruffe, presumably days to weeks from spawning, was as high as 15.8. Oocyte size was a better indicator of spawning readiness, with fish ready to spawn having oocyte diameters of 875–1005 μm, while the diameters of most stage 3 oocytes were <850 μm. The GSI was, however, adequate for showing that there

© 1997 NRC Canada



© 1997 NRC Canada

Fig. 4. Numbers of oocytes in various stages present in a typical cross section of an age 3+ ruffe ovary on different dates. (*a*) Prespawning ovary, April 10 sample, with numerous oocytes in several stages of development. (*b*) Spawning-ready ovary, May 2 sample, with other groups of oocytes in stage 3 and late stage 2. (*c*) Postspawning ovary, June 13 sample. This female had corpora lutea and many stage 2 and 3 oocytes in early stages of atresia; spawning appeared to have ceased and residual oocyte absorption to have begun. An asterisk denotes the presence of preovulatory atretic follicles.



was no second spawning period for DSH ruffe in July (or later). For example, the July 18 GSIs ranged from 0.4 to 1.0, and corresponding oocyte diameters were 130–320 μm (stage 2 oocytes). This was well below spawning values for either of these measurements.

The 1994 spawning period estimates based on the GSI (Selgeby 1995) and our histologically based estimates corresponded closely. However, histological data suggested that fewer age groups were involved in the actual spawning. The GSI data implied that some age 1 fish would spawn (Selgeby 1995). In our histological samples, even if an age 1 fish had an outlier GSI value, e.g., 9.7, its oocytes were as immature (stage 2) as fish with a low GSI. No histological evidence was found to suggest that age 1 females participated in spawning in 1994. Moreover, a high GSI in age 2 ruffe did not necessarily mean that they would spawn, as most of these females seemed to resorb their oocytes rather than to ovulate.

Finally, judging from the advanced state of maturity of ruffe ovaries in November in the DSH, it is plausible that, in habitats (and years) with shorter winters and longer growing seasons, or with lower densities of ruffe, additional age groups may participate in spawning. Also, the spawning period may be longer, or there may even be two distinct spawning periods.

Acknowledgements

Support for the present study was provided by Minnesota Sea Grant, the Minnesota Department of Natural Resources, the National Biological Service, Ashland, Wis., office, and the U.S. Fish and Wildlife Service, Ashland, Wis., office. We thank James Selgeby and the staff of the National Biological Service, Great Lakes Center Biological Station, Ashland, Wis., for providing fish for the present study. We also thank Dr. Edward Halls for his help on several aspects of this project.

References

Craig, J.F. 1987. The biology of perch and related fish. Timber Press, Portland, Ore.

deVlaming, V., Grossman, G., and Chapman, F. 1982. On the use of the gonosomatic index. Comp. Biochem. Physiol. A, **73**: 31–39.

Fairchild, D.J., and McCormick, J.H. 1996. Effects of temperature on hatching and development of ruffe (*Gymnocephalus cernuus*). J. Great Lakes Res. **22**: 89–94.

Jackson, L.E., and Sullivan, C.V. 1995. Reproduction in white perch: the annual gametogenic cycle. Trans. Am. Fish. Soc. **124**: 563–577.

Koonce, J.F., Bagenal, T.B., Carline, R.F., Hokanson, K.E.F., and Nagiec, M. 1977. Factors influencing year-class strength of percids: a summary and a model of temperature effects. J. Fish. Res. Board Can. **34**: 1900–1909.

Koshelev, B.V. 1963. Morpho-ecological peculiarities of oogenesis in related species of percoid fishes. Tr. Inst. Morfol. Zhivotn. Akad. Nauk SSSR, **38**: 189–231.

Kovalev, P.M. 1973. Natural reproductive conditions of the pike-perch (*Lucioperca lucioperca* L.), perch (*Perca fluviatilis* L.) and ruffe (*Aceina cernua* L.) of Lake Ilmen. J. Ichthyol. **13**: 943–946.

Kramer, R.H., and Smith, L.L., Jr. 1962. Formation of year classes in largemouth bass. Trans. Am. Fish. Soc. **91**: 29–41.

Leino, R.L., McCormick, J.H., and Jensen, K.M. 1990. Multiple effects of acid and aluminum on brood stock and progeny of

© 1997 NRC Canada

fathead minnows with emphasis on histopathology. Can. J. Zool. **68**: 234–244.

Neja, Z. 1988. On some problems of reproduction of ruff, *Gymnocephalus cernuus* (L., 1758) in the Lake Dabie. Acta Ichthyol. Piscatoria, **18**: 33–50.

Ogle, D.H. 1995. Ruffe, *Gymnocephalus cernuus*: a review of published literature. Administrative report No. 38. Wisconsin Department of Natural Resources Bureau of Fisheries Management, Madison, Wis.

Ogle, D.H., Selgeby, J.H., Savino, J.F., Newman, R.M., and Henry, M.G. 1996. Predation on ruffe by native fishes of the St. Louis River Estuary, Lake Superior, 1989–1991. N. Am. J. Fish. Manage. **16**: 115–123.

Pratt, D.M., Blust, W.H., and Selgeby, J.H. 1992. Ruffe, *Gymnocephalus cernuus*: newly introduced in North America. Can. J. Fish. Aquat. Sci. **49**: 1616–1618.

Selgeby, J.H. 1995. Abundance, distribution, biology, and community impacts of invading Eurasian ruffe. Final report to the Environmental Protection Agency No. 149-359-35-01-0. U.S. Environmental Protection Agency, Duluth, Minn.

Selman, K., and Wallace, R.A. 1986. Gametogenesis in *Fundulus heteroclitus*. Am. Zool. **26**: 173–192.

Treasurer, J.W., and Holliday, F.G.T. 1981. Some aspects of the reproductive biology of perch *Perca fluviatilis* L. A histological description of the reproductive cycle. J. Fish Biol. **18**: 359–376.

© 1997 NRC Canada

Echinogammarus ischnus: another crustacean invader in the Laurentian Great Lakes basin

Jonathan D.S. Witt, Paul D.N. Hebert, and William B. Morton

Abstract: We conducted a targeted analysis of the lower Great Lakes to determine the presence or absence of two euryhaline amphipod species, *Echinogammarus ischnus* Stebbing, 1899 and *Corophium curvispinum* (Sars, 1895), with an invasion history in Europe. *Echinogammarus ischnus* was identified for the first time in North America at a Detroit River site but was not detected at sites in lakes Erie and St. Clair. *Corophium curvispinum* was not detected at any of the sites. In contrast with earlier crustacean invaders, *E. ischnus* reproduces sexually and lacks a planktonic life stage. Allozyme analyses suggest that the Detroit River population was founded by sufficient individuals to maintain an average level of genetic variation.

Résumé : Nous avons réalisé une étude ciblée des Grands Lacs inférieurs pour déterminer la présence ou l'absence de deux espèces d'amphipodes euryhalins, *Echinogammarus ischnus* Stebbing 1899 et *Corophium curvispinum* (Sars, 1895), qui ont connu une explosion en Europe. *Echinogammarus ischnus* a été identifié pour la première fois en Amérique du Nord dans un site de la rivière Détroit, mais n'a pas été décelé dans les sites étudiés dans les lacs Érié et Saint-Clair. *Corophium curvispinum* n'a été décelé dans aucun des sites étudiés. Contrairement aux crustacés envahisseurs antérieurs, *E. ischnus* se reproduit sexuellement et il lui manque un stade de vie planctonique. Les analyses d'allozymes indiquent que la population de la rivière Détroit a été fondée par un nombre suffisant grand d'individus pour maintenir un niveau moyen de variation génétique.
[Traduit par la Rédaction]

Introduction

The Laurentian Great Lakes have become the subject of an extraordinary unplanned exercise in biomanipulation. Human activities, principally those related to shipping, have led to the recognized introductions of at least 60 animal species (Mills et al. 1993). Many of the Eurasian species among this group have invaded the Great Lakes since 1980. The relative paucity of such colonizations in the first 20 years following completion of the St. Lawrence Seaway suggests that the biological communities of the Great Lakes have recently become more permeable, or prone to colonization by exotic species, perhaps as a secondary consequence of the ongoing cascade of changes in species composition. Unfortunately, documentation of this shift in permeability is difficult because the occurrence and sequence of species arrivals are poorly understood. With the exception of morphologically distinctive taxa, few species are detected at the time of their arrival. In most cases, morphologically cryptic species can remain undetected for an extended time, especially in groups that are the subject of little taxonomic study. Resolution of this problem will require detailed studies of biological diversity in the various taxonomic groups constituting the Great Lakes fauna. However, as an initial focus for this work, it is important to examine the Great Lakes fauna for species recognized as invaders in other geographic regions, such as Europe.

Despite its importance in aquatic food webs, the crustacean fauna of the Great Lakes has received little attention. However, among the approximately 200 species of crustaceans known from the Great Lakes, 5 have been recognized as introductions from Eurasia, and 4 of these species are now common in the Great Lakes. The first reported invader, *Bosmina coregoni*, was detected in Lake Michigan in 1966 (Wells 1970) and now forms a numerically important component of the Great Lakes zooplankton fauna (Balcer et al. 1984). The mitten crab (*Eriocheir sinensis*) has also been detected in the Great Lakes on several occasions but has failed to establish reproducing populations (Nepszy and Leach 1973). The predatory cladoceran *Bythotrephes cederstroemi* was first reported in Lake Huron in 1984 (Bur et al. 1986), and is now a dominant component of the Great Lakes zooplankton. Finally, recent genetic studies have detected the occurrence of two other crustacean invaders that were overlooked by prior morphological studies. One of these species, *Bosmina maritima*, is now prevalent in the winter zooplankton community of the Great Lakes (De Melo and Hebert 1994), while hybrids derived from European *Daphnia galeata* now dominate the lower Great Lakes (Taylor and Hebert 1993). All four of the invading crustaceans that have established populations in the Great Lakes are planktonic organisms capable of parthenogenesis. Their planktonic lifestyle presumably facilitated their uptake and transfer in ballast water, while their parthenogenetic mode of reproduction would aid population establishment from a small number of founders. *Eriocheir sinensis* also possesses planktonic larval stages that likely facilitated its

Received July 25, 1996. Accepted October 8, 1996.
J13573

J.D.S. Witt,[1] P.D.N. Hebert, and W.B. Morton. Department of Zoology, University of Guelph, Guelph, ON N1G 2W1, Canada.

[1] Author to whom all correspondence should be addressed.
e-mail: jwitt@uoguelph.ca

© 1997 NRC Canada

introduction. Its failure to establish a reproducing population is probably a consequence of the zoea larva's intolerance to low salinity (Anger 1991).

Other crustacean species may have invaded the Great Lakes, but remain undetected because of limited taxonomic study. One such taxonomically neglected group, the Amphipoda, is known to include two euryhaline species, *Echinogammarus ischnus* Stebbing, 1899 and *Corophium curvispinum* (Sars, 1895), that have greatly expanded their European distributions as a result of canal construction and, in the latter case, ballast water transfers (Jazdzewski 1980; Kohn and Waterstraat 1990; Jazdzewski and Konopacka 1993; Van den Brink et al. 1993; Konopacka and Jesionowska 1995). These species, which are native to the Caspian – Black Sea region, show a close association with *Dreissena polymorpha* at some European localities, where the three invaders are suspected to have immigrated together (Kohn and Waterstraat 1990). *Corophium curvispinum*, which constructs and inhabits tubes made of mud, has reached extraordinarily high densities at some sites that it has invaded (Den Hartog et al. 1992; Van Den Brink et al. 1993).

This study reports the results of a survey on the lower Great Lakes to determine the presence or absence of these European amphipods. We demonstrate that one of these two species, *Echinogammarus ischnus* (synonym = *Chaetogammarus ischnus*), is now established in the Great Lakes basin. An electrophoretic study was also conducted to gain a sense of the size of the founding population.

Methods and materials

Twelve nearshore sites on lakes Erie and St. Clair and on the Detroit River were qualitatively sampled for amphipods between September 14 and 28, 1995, by sweeping a 350-μm mesh size dip net through vegetation, rocks, and debris at depths up to 1 m. The amphipods in these collections were examined for known members of the Great Lakes fauna using keys by Bousfield (1958), as well as for *Corophium* and *Echinogammarus* using keys in Barnard and Barnard (1983) and descriptions by Sars (1896) and Kohn and Waterstraat (1990). Following its discovery, specimens of *Echinogammarus ischnus* were preserved in 70% ethanol or flash frozen for electrophoretic work. Preserved specimens were then sorted into males, females, and subadults (on the basis of the presence or absence of papillae and oostegites) and measured from rostrum to telson using a digital system. Ovigerous females were enumerated and embryos were counted.

Total body extracts were prepared from the amphipods and levels of allozyme variation determined using cellulose acetate electrophoresis (Hebert and Beaton 1993) for 11 enzyme loci including two aldehyde oxidase (*Ao*) loci, arginine kinase (*Ark*), aspartate amino transferase (*Aat*), fumarate hydratase (*Fum*), isocitrate dehydrogenase (*Idh*), lactate dehydrogenase (*Ldh*), malate dehydrogenase (*Mdh*), malic enzyme (*Me*), phosphoglucomutase (*Pgm*), and glucose-6-phosphate isomerase (*Gpi*).

Results

Occurrence and identification

Only a single amphipod species was detected at each of the 12 sites (Fig. 1). At 11 of these localities, either *Hyalella azteca* or *Gammarus fasciatus* was present. *Corophium curvispinum* was not detected at any site, but *Echinogammarus ischnus* was detected at the sole Detroit River site. More than 400 *Echinogammarus ischnus* specimens were captured at this site beneath concrete slabs strewn over a sandy substrate at depths ranging from 0.05 to 1.0 m, and under wrack that had accumulated at the shoreline. It was not, however, collected from a nearby bed of *Valisneria* sp., or over the sand. A sample collected in mid-September included 93 females, 61 males, and 50 subadults. Seventy-three percent of the females were ovigerous with brood sizes ranging from 6 to 18 (n = 20, $\bar{x}$ = 11.6, SD = 3.3). The size of adult males ranged from 6.0 to 9.1 mm ($\bar{x}$ = 7.2 mm, SD = 0.07), while adult females ranged from 5.3 to 8.0 mm ($\bar{x}$ = 6.7 mm, SD = 0.07) and subadults ranged from 3.9 to 6.1 mm ($\bar{x}$ = 5.0 mm, SD = 0.07).

Fig. 1. Map showing localities of *Hyallela azteca* (triangles), *Gammarus fasciatus* (solid circles), and *Echinogammarus ischnus* (open circle).

Table 1. Allele frequencies and individual expected heterozygosities (H) at the two polymorphic loci of *Echinogammarus ischnus* in the Detroit River.

		Allele frequency		
Locus	n	Slow	Fast	H
Gpi	66	0.30	0.70	0.42
Pgm	58	0.22	0.78	0.35

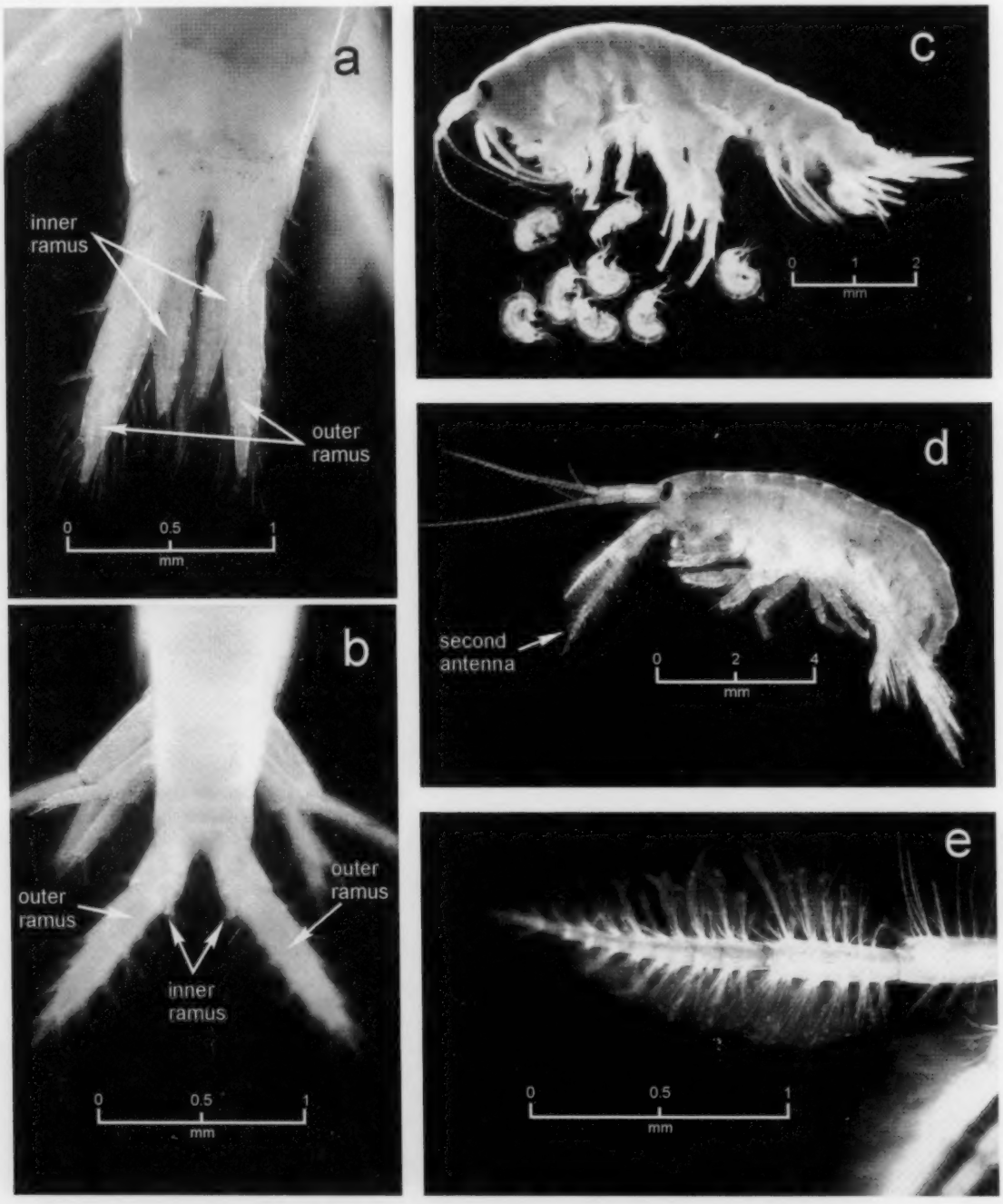
Current taxonomic keys that include the Great Lakes amphipod fauna (e.g., Bousfield 1958; Holsinger 1972; Pennak 1989) result in the placement of *Echinogammarus* in the genus *Gammarus*. However, these two genera can easily be separated by the striking difference in the morphology of their third uropods (Barnard and Barnard 1983). In *Gammarus*, the inner and outer rami are well developed (Fig. 2*a*), while the inner rami are vestigial in *Echinogammarus* (Fig. 2*b*). In *Echinogammarus ischnus*, the telson is cleft to the base with two dorsal and three apical spines on each lobe, distinguishing this species from the other members of its genus. *Echinogammarus ischnus* exhibits strong sexual dimorphism (Figs. 2*c* and 2*d*), with the male second antennae much more densely setose (Fig. 2*e*) than those of the female. Barnard and Barnard (1983) and Kohn and Waterstraat (1990) provide detailed descriptions of the genus *Echinogammarus* and *Echinogammarus ischnus*, respectively.

Electrophoresis

Three loci (*Ao*-1, *Aat*, and *Idh*) could not be resolved and are not further considered. Of the eight loci that resolved clearly,

© 1997 NRC Canada

Fig. 2. (*a*) *Gammarus* sp., third uropods; (*b*) *Echinogammarus ischnus*, third uropods; (*c*) *Echinogammarus ischnus*, female with brood; (*d*) *Echinogammarus*, male; (*e*) *Echinogammarus ischnus*, male second antenna.



only *Gpi* and *Pgm* were polymorphic, each with two alleles. The alleles were easily discriminated, with the faster alleles having relative mobilities of 1.15 and 1.19 at the *Gpi* and *Pgm* loci, respectively. Allozyme phenotypes of heterozygotes at these loci were concordant with those expected on the basis of the quaternary structures of the enzymes (Ward et al. 1992). Genotypic frequencies at both the *Gpi* and *Pgm* loci were in Hardy–Weinberg equilibrium ($\chi^2 = 2.7$ and 2.5, respectively, $p > 0.10$). Individual heterozygosities averaged over the eight loci were 9.6% (Table 1).

© 1997 NRC Canada

Discussion

This study has revealed the presence of a breeding population of the European amphipod *Echinogammarus ischnus* in the Detroit River. Genotypic frequencies at both polymorphic loci were concordant with Hardy–Weinberg expectations, confirming that the species reproduces sexually. The average heterozygosity for individuals of *Echinogammarus ischnus* from the Detroit River population is similar to those reported for other species of *Echinogammarus* (Lop and Oliver 1989), suggesting that the Detroit River population was founded by enough individuals to prevent a major loss of genetic variation.

The population consisted of adults and subadults with females carrying broods as large as 18 individuals. Prior work in Europe has shown that the life history of this amphipod is variable. Its life cycle has been studied most intensively in a thermally polluted lake in Poland, where breeding begins in February and peaks in June when 98% of females are ovigerous (Konopacka and Jesionowska 1995). During July, most females lack eggs, but a second burst of reproduction occurs in early fall before ceasing in November. Brood sizes in this population ranged from 2 to 27 and were related to female size. In Lake Kummerow in Germany, females produce broods of 20 or less during the summer, with reproduction terminating in late September (Kohn and Waterstraat 1990). The life history of *Echinogammarus ischnus* in its native Russia and Ukraine has been summarized by Konopacka and Jesionowska (1995). In the Dnieper River Delta in the Ukraine, reproduction takes place from mid-April until October but ceases from November to March. Brood sizes range from 4 to 41. In tributaries of the Don River in Russia, where the water temperature has a small annual range, *Echinogammarus ischnus* breeds throughout the year, producing broods of up to 48 individuals.

The range extension of *Echinogammarus ischnus* in Europe has been reviewed by Jazdzewski (1980). The species dispersed from the Caspian and Black Sea basins via man-made canals to the Baltic and North Sea basins. It was first recorded outside its native range in 1928, when it was discovered in the Vistula River, having entered via the Pripet'-Bug canal. By 1960, it had colonized the lower Neman River, gaining access via the Neman-Pripet' canal. In the 1970s, it was discovered in the Dortmund-Ems canal in Germany, which it reached via the extensive canal system linking European rivers. In the Danube River, it has advanced 1856 km upstream to Jarovce, Slovakia. Very recently, it has been discovered in The Netherlands, having immigrated from Germany via the Rhine River (Van den Brink et al. 1993). Clearly, *Echinogammarus ischnus* has the ability to disperse great distances along natural and man-made watercourses. As a result, its further colonization of the Great Lakes basin is likely to be rapid. It will most likely colonize the lower Great Lakes and their large tributaries and eastwards through the St. Lawrence River system. The species is also likely to colonize the large river systems of the United States such as the Mississippi and Missouri.

The impact of *Echinogammarus ischnus* on the indigenous fauna of the Great Lakes is difficult to predict. One of the best documented alien amphipod invasions is that of *Gammarus tigrinus*, a North American euryhaline species that has colonized The Netherlands, where it has displaced *Gammarus duebeni*, *Gammarus zaddachi*, and *Gammarus pulex* (Pinkster et al. 1977, 1992). The Detroit River population of *Echinogammarus ischnus* occupied a habitat typical of *G. fasciatus*, suggesting the possibility of competitive displacement of the latter species.

It is noteworthy that many recent Great Lakes invaders such as *Dreissena* spp., the gobies *Proterorhinus* and *Neogobius*, and now *Echinogammarus* are euryhaline, a likely consequence of the fact that most ocean-going vessels load ballast in estuarine settings. The colonization of the Great Lakes by *Echinogammarus* as well as *Neogobius* (Jude et al. 1992), via ballast water transfers, is interesting because these organisms lack both a pelagic stage and parthenogenetic reproduction, two traits that facilitate ballast water transfer and subsequent colonization. It is possible that these species represent a new wave of colonizations of the Great Lakes by sexually reproducing organisms that lack planktonic stages, as a result of an increase in permeability of the Great Lakes communities to invading organisms. This study is the first to establish the value of investigating the Great Lakes for organisms with a known invasion history in other geographic areas. Interestingly, *Corophium curvispinum*, which has a history of ballast water transfers in Europe, seems to have so far failed to colonize the Great Lakes, while *Echinogammarus ischnus* has succeeded despite a lack of evidence for this mode of dispersal in Europe. This result reinforces the "sweepstakes" nature of the colonization process. However, given the invasion history of *Corophium*, its colonization of the Great Lakes seems probable. The results of this study make it apparent that more intensive studies of the Great Lakes are required to detect and document the occurrence of other exotic species, especially in taxonomically neglected groups.

Acknowledgements

We thank Dr. E.L. Bousfield for confirming our identification and R. Geddes and I. Smith for aid with the figures. Dr. A. Loche's comments improved an earlier version of the manuscript. Funding for this research program was provided by grants from the Natural Sciences and Engineering Research Council and the Max Bell Foundation to P.D.N.H.

References

Anger, K. 1991. Effects of temperature and salinity on the larval development of the Chinese mitten crab *Eriocheir sinensis* (Decapoda: Grapsidae). Mar. Ecol. Prog. Ser. **72**:103–110.

Balcer, M.D., Korda, N.L., and Dodson, S.I. 1984. Zooplankton of the Great Lakes. A guide to the identification of the common crustacean species. University of Wisconsin Press, Madison, Wis.

Barnard, J.L., and Barnard, C.M. 1983. Freshwater Amphipoda of the world. Vols. 1 and 2. Hayfield Assoc., Mt. Vernon, Va.

Bousfield, E.L. 1958. Fresh-water amphipod crustaceans of glaciated North America. Can. Field-Nat. **72**: 55–113.

Bur, M.T., Klarer, D.M., and Krieger, K.A. 1986. First records of a European cladoceran, *Bythotrephes cederstroemi*, in lakes Huron and Erie. J. Great Lakes Res. **12**: 144–146.

De Melo, R., and Hebert, P.D.N. 1994. A taxonomic reevaluation of North American Bosminidae. Can. J. Zool. **72**: 1808–1825.

Den Hartog, C., Van Den Brink, F.W.B., and Van Der Veld, G. 1992. Why was the invasion of the river Rhine by *Corophium curvispinum* and *Corbicula* species so successful? J. Nat. Hist. **26**: 1121–1129.

© 1997 NRC Canada

Hebert, P.D.N., and Beaton, M. 1993. Methodologies for allozyme analyses using cellulose acetate electrophoresis. Helena Laboratories, Beaumont, Tex.

Holsinger, J.R. 1972. The freshwater amphipod crustaceans (Gammaridae) of North America. Biota of freshwater ecosystems identification manual No. 5. U.S. Environmental Protection Agency, Washington, D.C.

Jazdzewski, K. 1980. Range extensions of some gammaridean species in European inland waters caused by human activity. Crustaceana (Leiden), **6**(Suppl.): 84–107.

Jazdzewski, K., and Konopacka, A. 1993. Survey and distribution of Crustacea Malacostraca in Poland. Crustaceana (Leiden), **65**: 176–191.

Jude, D.J., Reider, R.H., and Smith, G.R. 1992. Establishment of Gobiidae in the Great Lakes basin. Can. J. Fish. Aquat. Sci. **49**: 416–421.

Kohn, J., and Waterstraat, A. 1990. The amphipod fauna of Lake Kummerow (Mecklenburg, German Democratic Republic) with reference to *Echinogammarus ischnus* Stebbing, 1899. Crustaceana, **58**: 74–82.

Konopacka, A., and Jesionowska, K. 1995. Life history of *Echinogammarus ischnus* (Stebbing, 1898) (Amphipoda) from artificially heated Lichenskie Lake. Crustaceana, **68**: 341–349.

Lop, A.F., and Oliver, J.L. 1989. Isozyme differentiation among sibling species and among populations of the *Echinogammarus berilloni* group (Crustacea, Amphipoda). Z. Zool Syst. Evolutionsforsch. **27**: 282–296.

Mills, E.L., Leach, J.H., Carlton, J.T., and Secor, C.L. 1993. Exotic species in the Great Lakes: a history of biotic crises and anthropogenic introductions. J. Great Lakes Res. **19**: 1–54.

Nepszy, S.J., and Leach, J.H. 1973. First records of the Chinese mitten crab, *Eriocheir sinensis*, (Crustacea: Brachyura) from North America. J. Fish. Res. Board Can. **30**: 1909–1910.

Pennak, R. 1989. Freshwater invertebrates of the United States. Wiley, New York.

Pinkster, S., Smit, H., and Brandse-de Jong, N. 1977. The introduction of the alien amphipod *Gammarus tigrinus* Sexton, 1939, in the Netherlands and its competition with indigenous species. Crustaceana (Leiden), **4**(Suppl.): 91–105.

Pinkster, S., Scheepmaker, N., Platvoet, D., and Broodbakker, N. 1992. Drastic changes in the amphipod fauna (Crustacea) of Dutch inland waters during the last 25 years. Bijdr. Dierkd. **61**: 193–204.

Sars, G.O. 1896. Crustacea Caspia. Contributions to the knowlege of the carcinological fauna of the Caspian Sea. Amphipoda. Bull. Acad. Imp. Sci. St. Petersbourg, **6**(Suppl.): 233–264.

Taylor, D.J., and Hebert, P.D.N. 1993. Cryptic intercontinental hybridization in *Daphnia* (Crustacea): the ghost of invasions past. Proc. R. Soc. Lond. B Biol. Sci. **254**: 163–168.

Van den Brink, F.W.B., Paffen, B.G.P., Oesterbroek, G., and Van der Velde, G. 1993. Immigration of *Echinogammarus* (Stebbing, 1906) (Crustacea, Amphipoda) into the Netherlands via the lower Rhine. *In* Bulletin Zoologisch Museum, Universiteit van Amsterdam. Vol. 13. Institut of Taxonomic Zoology, University of Amsterdam, Amsterdam, The Netherlands. pp. 167–170.

Ward, R.D., Skibinski, D.O.F., and Woodwark, M. 1992. Protein heterozygosity, protein structure, and taxonomic differentiation. Evol. Biol. **26**: 73–159.

Wells, L. 1970. Effects of alewife predation on zooplankton populations in Lake Michigan. Limnol. Oceanogr. **15**: 556–565.

© 1997 NRC Canada

Effects of fish density and river fertilization on algal standing stocks, invertebrate communities, and fish production in an arctic river

Linda A. Deegan, Bruce J. Peterson, Heidi Golden, Carole C. McIvor, and Michael C. Miller

Abstract: This study examined the relative importance of bottom-up and top-down controls of an arctic stream food web by simultaneous manipulation of the top predator and nutrient availability. We created a two-step trophic system (algae to insects) by removal of the top predator (Arctic grayling, *Thymallus arcticus*) in fertilized and control stream reaches. Fish abundance was also increased 10 times to examine the effect of high fish density on stream ecosystem dynamics and fish. We measured the response of epilithic algae, benthic and drifting insects, and fish to nutrient enrichment and to changes in fish density. Insect grazers had little effect on algae and fish had little effect on insects. In both the control and fertilized reaches, fish growth, energy storage, and reproductive response of females declined with increased fish density. Fish growth and energy storage were more closely correlated with per capita insect availability than with per capita algal standing stock.

Résumé : La présente étude porte sur l'importance relative des contrôles ascendants et descendants dans un réseau alimentaire d'un cours d'eau de l'arctique par la manipulation simultanée du prédateur suprême et de la disponibilité des éléments nutritifs. Nous avons créé un système trophique à deux étages (algues à insectes) en éliminant le prédateur suprême (ombre de l'Arctique, *Thymallus arcticus*) dans des tronçons fertilisés et témoins du cours d'eau. L'abondance des poissons a également été multipliée par un facteur de 10 pour examiner l'effet d'une forte densité de poissons sur les caractéristiques dynamiques de l'écosystème du cours d'eau et sur le poisson. Nous avons mesuré la réponse des algues épilithiques, des insectes benthiques et dérivants, et des poissons à la fertilisation du cours d'eau et aux variations de la densité du poisson. Les brouteurs d'insectes avaient peu d'effets sur les algues et le poisson avait peu d'effet sur les insectes. Chez les poissons présents dans les deux tronçons, fertilisé et témoin, la croissance et le stockage d'énergie ainsi que la réponse reproductive des femelles ont diminué avec l'augmentation de la densité de poissons. La croissance et le stockage d'énergie chez les poissons étaient corrélés plus étroitement avec la disponibilité des insectes par tête qu'avec la biomasse d'algues par tête.
[Traduit par la Rédaction]

Introduction

Understanding the relative importance of top-down biotic versus bottom-up (abiotic) controls is central to understanding and predicting ecosystem dynamics and community composition. The balance between top-down and bottom-up control on a given trophic level is hypothesized to depend on both primary production and the number of trophic levels (Oksanen et al. 1981; Fretwell 1987; Oksanen 1990; Abrams 1993). There is also evidence for dual control in some lake systems (Carpenter et al. 1987, 1990; Persson et al. 1992). Early studies in streams indicated that accumulation of algal biomass was bottom-up controlled by either nutrients or light (Stockner and Shortreed 1978; Elwood et al. 1981; Peterson et al. 1983); however, recent work has found herbivory (top-down) to be an important factor (Matthews et al. 1987; Stewart 1987; Power 1990; Gelwick and Matthews 1992; Peterson et al. 1993*a*). Dual control is possible in streams, but few studies (e.g., Rosemond et al. 1993) have simultaneously tested the relative importance of top-down – bottom-up controls on primary production and none has simultaneously measured the impacts on the top predator.

The purpose of this study was to examine the relative importance of bottom-up and top-down controls of food webs by simultaneous manipulation of nutrient availability by fertilization and top predator density in an arctic stream. For several reasons, we might expect that top-down control could be more important in arctic than in temperate streams. Once the effect of nutrient limitation is removed by fertilization, we would expect algal and insect populations to increase. In addition, unlike the situation in temperate rivers, drifting insects have no refuge from predation in darkness in the Arctic (Hinterleitner-Anderson et al. 1992), and Arctic grayling (*Thymallus arcticus*) are known to feed for 24 h (L.A. Deegan, personal observation). Thus, fish have more opportunity to control the abundance of the important insect grazers in the

Received November 22, 1994. Accepted July 16, 1996.
J12641

L.A. Deegan, B.J. Peterson, and H. Golden. The Ecosystems Center, Marine Biological Laboratory, Woods Hole, MA 02543, U.S.A.
C.C. McIvor. University of Arizona, Arizona Cooperative Fish and Wildlife Research Unit, School of Renewable Natural Resources, Tucson, AZ 85721, U.S.A.
M.C. Miller. University of Cincinnati, Biological Sciences, Cincinnati, OH 45221-0006, U.S.A.

© 1997 NRC Canada

Arctic compared with temperate regions. In this study, we take advantage of a long-term river fertilization project to examine the interactive effects of changes in river productivity and the abundance of the top predator, Arctic grayling, on stream ecosystem dynamics and fish production. We evaluate the effect of these interactions starting at the base of the food web with nutrient additions through primary producers (epilithic algae), insects, and the response of the fish.

The experiment was designed with two levels of nutrient availability (control and fertilized) and three levels of fish abundance (no fish, average, and high). We created a two-step trophic system (algae to insects) by removal of the top predator (fish) in control and fertilized stream reaches. In addition, fish abundance was increased 10 times average to examine the effects of high fish density on stream ecosystem dynamics and on the top predator. If top-down control is operating in an odd-numbered trophic level system (three levels), Fretwell and Oksanen's model (Oksanen et al. 1981; Fretwell 1987; Oksanen 1990) predicts that removal of the top level (fish) should release the grazers (insects) from predation control, resulting in algal biomass decline. Conversely, an increase in predation pressure on the insect grazers should result in the accumulation of algal biomass. The top level (fish) is predicted to be limited by competition for food.

Background and site description

The Kuparuk River (68°38′N, 149°24′W) on the North Slope of Alaska provided a unique opportunity to test these ideas because of its relatively simple food web and long-term nutrient addition experiment (Peterson et al. 1993*a*). The ecology and biogeochemistry of the Kuparuk River, which is a clear-water tundra stream, are described by Peterson et al. (1985, 1986, 1993*a*), Lock et al. (1989), and Kriet et al. (1992). P and in some years N have been added to a reach of the Kuparuk River near the Dalton Highway every year from 1983 through 1994 (Deegan and Peterson 1992; Peterson et al. 1993*a*). The experimental reach of the river alternates between pools and riffles, with a substratum of rocky cobble and boulders. Riffle sections average about 30 m long and 10 m wide (300 m^2), and at mean summer flows, water is 5–30 cm deep. Pools range between 1 and 2 m in depth in midsummer and are 10–50 m long and 10–20 m wide. The Kuparuk River is frozen solid from October until May or June and has a high discharge in June as the result of snowmelt. River flow is lower in midsummer, although it responds quickly to rain or snow events because permafrost prevents deep seepage. In August, light and temperature decline sharply and the river freezes again in late September or October.

The Kuparuk River is a three-level trophic system consisting of relatively few species. The first trophic level is epilithic (a microbial and algal coating on rocks) production, which is an important food source to both grazers and filter feeders (Peterson et al. 1993*b*). The insects that make up the second trophic level include blackfly larvae (Simuliidae), which are filter feeders, mayfly nymphs (Baetidae), which graze on the epilithon, caddisfly larvae (Brachycentridae), which are predominantly filter feeders but also graze on the epilithon, and chironomids (Chironomidae), which are both grazers and detritivores. The third trophic level consists of adult and young-of-the-year (YOY) Arctic grayling, the only fish species in this river reach (Deegan and Peterson 1992; Peterson et al. 1993*a*). Arctic grayling feed primarily on nymphs and emerging adults of mayflies and caddisflies (Reed 1964; Peterson et al. 1993*b*). Chironomids account for less than 10% of the diet. The abundance of adults is usually 50–100 fish per kilometre of river (three to six adults per pool), while the abundance of YOY Arctic grayling is highly variable from year to year (Deegan and Peterson 1992).

P fertilization of the Kuparuk River for four consecutive summers dramatically altered the controls on primary producers from bottom-up control under oligotrophic conditions to top-down control under eutrophic conditions (Peterson et al. 1993*a*). The sequence of responses that we measured over 1983–1987 is as follows. Initially, added phosphate stimulated the growth of epilithic algae. Both algal biomass and productivity increased and chlorophyll accumulated on the river bottom in the first two (1983, 1984) summers. Diatom community composition changed little in spite of large chlorophyll changes. In 1985, 1986, and 1987, however, there was little chlorophyll *a* accumulation as a result of P addition. On the premise that N might have become limiting as a result of the addition of surplus P, both P and P + N were added to the river in separate reaches in 1986. Although P + N increased epilithic chlorophyll *a* accumulation over control and P alone, the increase was small, indicating that N was not limiting. The increases in algal and bacterial production provided increased high-quality food for filtering and grazing insects (Peterson et al. 1993*a*). The insects responded with increased growth and, in the case of caddisflies and possibly mayflies, with increases in density (Hershey et al. 1988; Hiltner and Hershey 1992; Hinterleitner-Anderson et al. 1992). Blackfly density, however, declined because of competitive interaction with *Brachycentrus* and increased algal cover (Hiltner and Hershey 1992). Experimental evidence from bioassays using insecticides to deter grazers indicated that grazing insects (mayflies and chironomids) could control algal biomass accumulations (Gibeau and Miller 1989). Thus, the lack of chlorophyll *a* accumulation in the fertilized reach was probably due to grazing by insects in the 3rd to 5th years.

The fertilization also increased fish growth (Deegan and Peterson 1992). Both YOY and adult Arctic grayling grew faster and achieved better condition in the fertilized reach in response to the increases in insect size and abundance (Deegan and Peterson 1992). Adults increased neutral lipid storage (important for overwinter survival) and possibly reproductive potential (Deegan and Peterson 1992) by the end of the summer. YOY Arctic grayling in some years were twice as large in the fertilized reach compared with fish in the control reach by the end of the summer. We hypothesized that increased fish growth, survival, and reproduction could lead to top-down control on insects by fish.

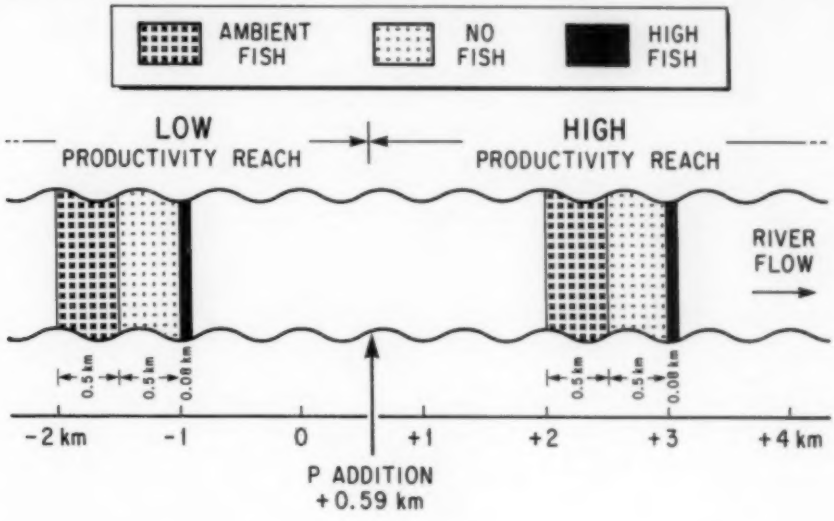
Methods

Whole-river nutrient addition

To increase river productivity, we added 10 μg $P \cdot L^{-1}$ as phosphoric acid during July 1 to August 15, 1988 (Fig. 1). Locations on the river were designated as plus (fertilized reach) or minus (control) kilometres in reference to the 1983 P addition site. In 1985, the point of P addition was moved to 0.59 km downstream; however, we have maintained the station location kilometre designations relative to the

© 1997 NRC Canada

Fig. 1. Experimental design of the river fertilization and the manipulation of the densities of the top trophic level, Arctic grayling.



original site to be consistent with other publications. The period July 1 to August 15 was chosen as the fertilization period because it spans the time of warmest waters and highest activity at all trophic levels during the brief Arctic summer.

Manipulation of fish density

We manipulated adult fish abundance by creating three fish densities in both the control and fertilized stream reaches (no fish, average, and high fish densities). After the fish distributed themselves into summer feeding territories in early July, we isolated three segments within both control and fertilized reaches by installing black plastic fish weirs (mesh size 2.5 cm, Deegan and Peterson 1992). This mesh size limits the movements of fish larger than 20 cm total length. Fish density treatments were arranged with the average fish abundance upstream (~500-m-long segment), no fish in the middle (~500-m segment), and high fish abundance downstream (~80-m segment) (Fig. 1). We felt that this arrangement of treatments would minimize any downstream effects of fish predation on drifting insect communities and provide a conservative test of the effects of manipulating fish abundance.

Because we were limited by the length of the fertilized reach and the length of each stream segment, we had only one fish density treatment per river reach (control or fertilized). Our previous experience indicated that we needed a sample of roughly 20 adult fish to detect a difference in growth response (Deegan and Peterson 1992). The minimum river length that would contain 20 fish at average fish density was about 500 m or five or six riffle/pool segments. We made the fish removal section 500 m also; however, the high fish density treatment was only 80 m (one riffle/pool section). The high fish density segment was shorter because obtaining enough fish to stock a 500-m section of the river to 10 times average density required removing fish from 5 km of river, which was not possible. We chose a density treatment of 10 times natural because we wanted to mimic a long-term increase in fish abundance and therefore needed a density manipulation that was greater than the year-to-year variability (0.5–5 times) in fish abundance. Average abundance of adult Arctic grayling, based on recaptures during the experiment as well as above- and below-water visual observations, was 50 fish·km^{-1} in 1988 but has ranged from 20 to 200 fish·km^{-1} over the years (Deegan and Peterson 1992). All fish captured in the no-fish segment and additional fish captured outside the three segments were added to the high fish abundance treatment to achieve roughly 10 times average density (~500 fish·km^{-1}). After the experiment was set up, we surveyed the treatments by snorkeling and saw only one or two fish in the fish-removal treatments, four or five untagged fish in the average-density treatment, and no untagged fish in the high-density treatments.

On August 3, a flood event topped all the weirs and flattened the weir at the downstream end of the high fish density treatment in the fertilized reach. This weir remained fully down for 18 h. During this time, at least 100 fish from downstream that were migrating upstream to overwinter entered and remained trapped in this treatment until the end of the experiment. Weirs were repaired and were again impassable to fish by August 5. Fish were recaptured from August 7 to 13, usually still in the segments in which they were originally placed; however, a few had moved upstream.

Stream flow and dissolved nutrients

Stream height was monitored with a continuously recording pressure sensor and converted to discharge by constructing a calibration curve from a series of discharge measurements (Kriet et al. 1992). Temperature was monitored by a temperature sensor and daily readings from max–min thermometers. Nutrient levels were monitored weekly by collecting water in 1-L dark polyethylene bottles at 12 stations from 2 km upstream of the nutrient addition site to 5 km downstream. Soluble reactive phosphate (SRP), nitrate plus nitrite, and ammonia were measured by semiautomated colorimetric methods (Wetzel and Likens 1979).

Epilithic response to river fertilization

To assess the response of epilithic algae to nutrient enrichment, we measured epilithic chlorophyll along a transect from the control to fertilized reach. Epilithic chlorophyll on riffle rocks was assayed by scrubbing a known area (0.78-cm^2 patch) of a rock (n = 5 rocks) into a known volume of water. A subsample of the water was extracted in 95% acetone at 4°C for 24 h. The extracted chlorophyll concentration was determined on a Turner 111 fluorometer (Strickland and Parsons 1972) and chlorophyll a was expressed as micrograms chlorophyll a per square centimetre.

© 1997 NRC Canada

Table 1. Initial fish density and mean size (±SE) of fish in the average and high fish density treatments in control and fertilized river reaches.

	River fertilization			
	Control		Fertilized	
	Average	High	Average	High
Number of fish tagged	20	43	29	45
Measured distance of treatment (km)	0.45	0.08	0.57	0.08
Fish density				
Fish·km^{-1}	44.4	537.75	50.8	562.5
Fish per pool/riffle (1000·m^{-2})	2.9	35.8	3.4	37.5
Total length (cm)	36.54	36.38	36.98	36.66
	(0.47)	(0.36)	(0.28)	(0.38)
Total weight (g)	401.55	397.70	411.66	395.98
	(13.07)	(10.58)	(15.89)	(12.05)
Condition factor	0.82	0.82	0.81	0.80
	(0.01)	(0.01)	(0.03)	(0.01)

Chlorophyll *a* bioassay to evaluate algal response to nutrient additions and insect exclusion

A bioassay consisting of agar chemical diffusing disks was used to evaluate the response of the epilithon to nutrients and insect grazing (Gibeau and Miller 1989; Peterson et al. 1993*a*). Each disk was composed of a 40-mL vial filled with various nutrient-supplemented agar treatments and sealed with a porous porcelain disk. Three vials were used for each disk treatment. Disk treatments were control (C, ultrapure agar with no chemical addition), malathion (M, 3.97 mL of commercial-grade malathion per litre of 2% agar), N + P (NP, 0.05 mol $NH_4Cl \cdot L^{-1}$ and 0.005 mol $K_2HPO_4 \cdot L^{-1}$), and N + P + malathion (NPM, 0.05 mol $NH_4Cl \cdot L^{-1}$, 0.005 mol $K_2HPO_4 \cdot L^{-1}$, and 3.97 mL malathion·L^{-1}). N and P were added to test for nutrient limitation and malathion was used to deter insects to test for grazing insect effects. Malathion is an insecticide that discourages insects from remaining on the disks (M.C. Miller, personal observation). We incubated the bioassays in riffles in each fish density treatment for 2 wk in July. To measure chlorophyll *a*, an entire disk was submerged in acetone for 24 h and the extracted chlorophyll *a* concentration was determined as for rocks.

Response of the insect community to river fertilization and fish density

To assess the response of the insect community to river fertilization and fish density manipulations, we measured insect standing stocks and drift. Density of invertebrates on riffle bottoms was estimated in the fertilized (July 15) and control (July 14) reaches. The timing of this sample was designed to estimate the levels of insects on the riffle rocks before emergence began (Hinterleitner-Anderson et al. 1992). Three samples, each consisting of four riffle rocks placed in a plastic basin (average area of 400 cm^2, Hershey and Hiltner 1988), were taken in each treatment. Insects were scrubbed off rocks with a soft-bristle brush. They were concentrated with 150-μm-mesh net and preserved in 95% ethanol.

Drift samples were taken in midafternoon in riffles in the control (July 14 and 27) and fertilized (July 15 and 28) reaches. Drift samples taken over a fairly short period (15–30 min) are representative of drift over longer time frames in the Kuparuk River because there is no diurnal variation in insect drift in July and early August (Hinterleitner-Anderson et al. 1992). During early July, paired 15-min samples were taken with drift nets (0.14-m^2 mouth opening, 350-μm mesh) set in the middle of each fish density treatment in the two productivity reaches. For the late July set, we used the same methods and locations but increased the sample time to 30 min because of low water current velocities. In early July, we sampled between 15 and 25 m^3 and in late July between 40 and 60 m^3 per drift sample. Drift samples were concentrated through 150-μm-mesh net and preserved in 95% ethanol. Water current readings were taken at the net opening with a Marsh McBirney EMF type current meter. The number of individuals per 100 m^3 was estimated from the current meter reading and the net area submerged.

Response of fish to river fertilization and fish density

Adult fish growth was measured by individually marking fish in the average and high fish density treatments (Table 1). Fish were anesthetized with 2-phenoxyethanol and total length (±0.1 cm) and weight (±1 g) were measured after fish were held in cages for 24 h. Fish were then tagged using individually numbered Floy tags and released. One-way ANOVAs using the combination of river productivity (control or fertilized) and fish density (average or high) as the main factor (four levels) indicated no difference in initial length ($F_{3,133} = 0.423$, $P = 0.73$), weight ($F_{3,133} = 0.285$, $P = 0.83$), or condition factor ($F_{3,133} = 0.78$, $P = 0.50$) of fish among treatments (Table 1). In mid-August, tagged fish were recaptured and measured. Changes in total weight, total length, and Fulton's condition factor (Bolger and Connolly 1989) were calculated as the difference between initial and final values for individually tagged fish (Deegan and Peterson 1992).

Long-term energy storage was estimated as weight of neutral lipid surrounding the intestine using hexane extraction and short-term energy storage as the hepatosomatic index (HSI) (Gardiner and Geddes 1980; Ohman 1988; Deegan and Peterson 1992). Reproductive response was measured as total wet mass of gonad tissue (Deegan and Peterson 1992). Organs were placed in separate preweighed containers, weighed (±0.001 g), and dried to constant weight at 60°C (usually 48 h). The containers were removed from the oven and placed immediately in a desiccator to cool and then dry weights were determined.

Data analysis

We sacrificed some statistical power to test for ecosystem and top trophic level effects in experimental units sufficiently large (river reaches) that normal behavior of the organisms and natural environmental conditions were assured. The combination of nutrient addition

© 1997 NRC Canada

(control or fertilized) and fish density was considered as the experimental treatment. Because of practical and ecological considerations, this experiment lacks true replication in the sense that each level of our treatment is associated with a specific site within a single river, a problem common to many large-scale field experiments (Carpenter 1990; Eberhardt and Thomas 1991). The nature of ecosystem-level experiments often results in low power to reject a false null hypothesis of "no difference" because of the inevitable constraints of limited or no replication (Winer 1971; Toft and Shea 1983; Hurlbert 1984; Peterman 1990; Johnson and Field 1993). We used multiple observations of each response variable (algal standing stock, insect densities, fish growth, etc.) to estimate treatment means (six treatments). Accordingly, we cannot state categorically that differences between treatments were the result of our manipulations, only that differences occurred. If differences were found, we examined these differences for consistency and used our history of work on this river (Peterson et al. 1993*a*; Deegan and Peterson 1992) as a measure of variability not associated with our treatment.

ANOVA was used to examine differences among treatments. For most analyses, we used a one-way ANOVA using the combination of fish density and river productivity as the main factor. For variables measured on several dates (such as chlorophyll or insect drift), we used repeated measures ANOVA. The bioassay data were analyzed using a two-way ANOVA using the combination of fish density and river productivity as one factor and the chemical treatment of the disk as the other main factor. The whole-river response of epilithon to river fertilization was tested with a one-way repeated measures ANOVA using river fertilization as the main factor and postfertilization measurements of chlorophyll *a* as the repeated variable. To balance type I and type II errors, we set $\alpha = 0.05$ and did an experimentwise adjustment of the error (Bonferroni correction) to control for the effects of running several ANOVAs on related data (Rice 1988; Huang and Sih 1990; Gelwick and Matthews 1992). Contrasts were used to test a priori hypotheses. Data were checked for normality (Wilks–Shapiro test) and heterogeneity of variance (Bartlett's test) and transformed when necessary. Insect densities (drift and benthic) and chlorophyll were $\ln(x + 1)$ transformed to correct for the positive correlation of variance and mean.

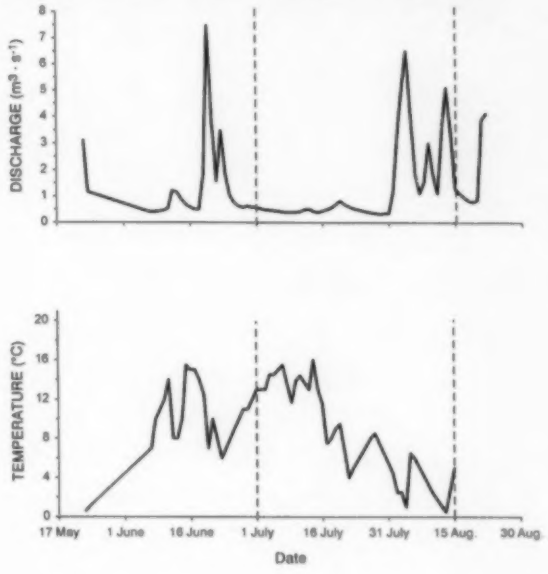
Results

Stream flow and nutrients

River discharge was exceptionally low and water temperature high during most of the experimental period (Fig. 2). Discharge was less than 2 $m^3 \cdot s^{-1}$ for most of the spring with a single 2-day peak in late June of 7 $m^3 \cdot s^{-1}$. Spring discharge values of 10–30 $m^3 \cdot s^{-1}$ which may last for several days, are common in this system (Peterson et al. 1993*a*). The low spring discharge was followed by 4–5 weeks of continuous, exceptionally low flows (<1 $m^3 \cdot s^{-1}$) until the August 3 rainstorm (peak river discharge during storm of 8 $m^3 \cdot s^{-1}$). Water temperature was also exceptionally high (12–16°C) in July and slowly declined over August because of declining day length and cloudy weather.

P addition increased SRP and caused slight declines in nitrate concentrations in stream water (Fig. 3). The average concentrations of nutrients in the upstream control reaches were low for SRP (0.1 μm) and ammonium (1.0 μm) and slightly higher for nitrate (2.0 μm) (Fig. 3). Phosphate concentrations increased 10-fold immediately downstream of the nutrient dripper (to 1.0 μm) and then declined over the next several kilometres. At the low flow typical of most of the experimental period, the added P was taken up within 5 km (Fig. 3). Ammonium levels were not changed by the P addition; however, nitrate levels declined as a result of uptake by epilithon in the fertilized reach (Fig. 3).

Fig. 2. Record of stream discharge and temperature during the 1988 field season. Vertical lines indicate the duration (July 1 to August 15) of the nutrient addition and fish manipulation experiments.



Epilithon response to river fertilization

Chlorophyll increased fivefold by the end of the summer in response to whole-river P addition (Fig. 4). By August 15, chlorophyll *a* levels exceeded 15 $\mu g \cdot cm^{-2}$ in the fertilized reach, and chlorophyll levels in the control reach were around 2 $\mu g \cdot cm^{-2}$ (repeated measures ANOVA, $F_{1,43} = 20.66$, $P = 0.0001$.) Algal biomass in the control reach was high in 1988 compared with other years (1983–1987 average = 0.5 $\mu g \cdot cm^{-2}$, Peterson et al. 1993*a*), indicating favorable environmental conditions for algal biomass accrual.

Effects of river fertilization and fish predation on insects

Total benthic invertebrate density in mid-July was around 10 000 individuals$\cdot m^{-2}$. Total density did not differ among treatments, although there were some differences in response among the insect groups (Fig. 5; Table 2). Small chironomids dominated the benthic community (roughly 50–60% numerically) in both the control and fertilized reaches. Blackflies (Simuliidae) were second (25–40%), followed by mayflies (Ephemeroptera, 20%) and caddisflies (Tricoptera, 1%). Consistent with previous years' observations, caddisfly abundance was higher (contrast analysis $F = 36.31$, $P < 0.05$) in the fertilized compared with the control reach (Hiltner and Hershey 1992; Peterson et al. 1993*a*). Chironomid, blackfly, and mayfly benthic abundances were similar between the two river reaches. Fish density had little effect on insect abundance.

Total invertebrate drift did not differ among treatments or over time, although drift of some insect groups did vary over time (Fig. 6; Table 2). The major components of the drift were

© 1997 NRC Canada

Fig. 3. Transects along the stream of average concentrations of phosphate, ammonium, and nitrate in the Kuparuk River postfertilization. The arrow at 0.59 km indicates the point of P addition. Samples to the left of the arrow represent the control reach; samples to the right indicate the fertilized reach.

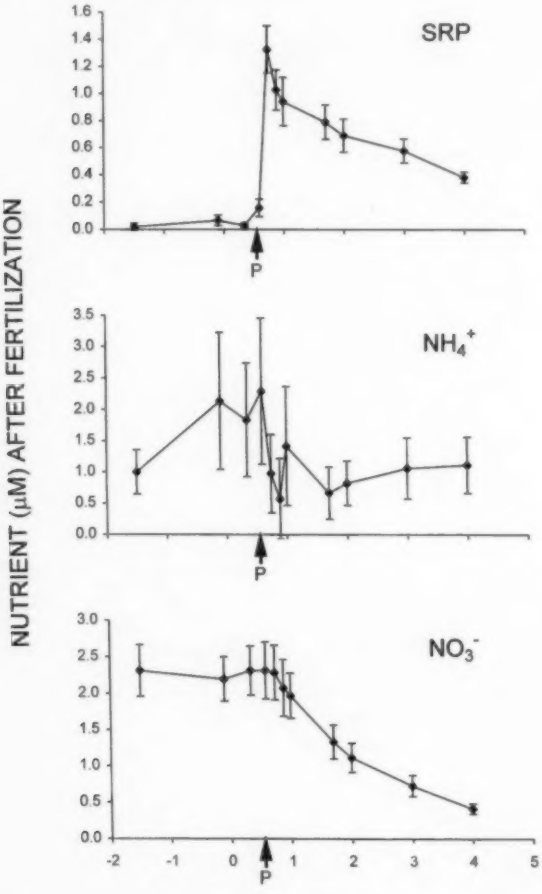
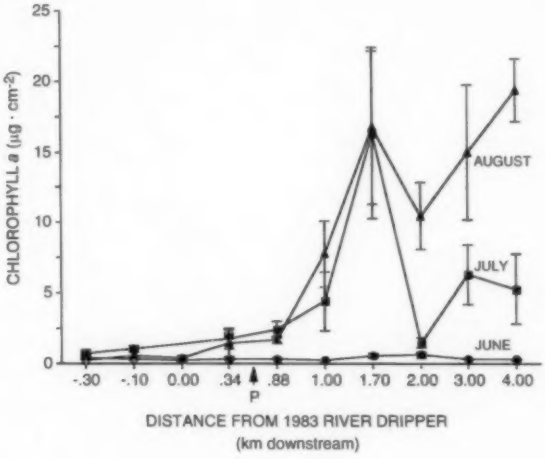


Fig. 4. Transects of epilithic algae (chlorophyll *a*) (±1 SE) along the whole-stream P enrichment in the Kuparuk River. The arrow at 0.59 km indicates the point of P addition. Samples to the left of the arrow represent the control reach; samples to the right indicate the fertilized reach. Repeated measures ANOVA on postfertilization (July 23 and August 15) chlorophyll *a* indicates a significant difference between the control and fertilized reaches ($F_{1,43} = 20.6$, $P = 0.0001$).



chironomids (40–50%), mayflies (25–40%), blackflies (10%), and caddisflies (5%). Within each time period, both mayfly and blackfly drift were lower in the fertilized reach compared with the control (Fig. 6). Chironomid and Tricoptera drift densities were not different between control and fertilized reaches (Fig. 6; Table 2). We also found no effect of fish density on drift abundance in either the control reach or the fertilized reach (Fig. 6; Table 2). Consistent with our expectations, fish density had no effect on chironomids and blackflies in the drift (Fig. 6). Contrary to our expectations, fish density had no effect on caddisfly or mayfly drift abundance (Fig. 6).

Response of chlorophyll *a* bioassay to added nutrients and insect grazing exclusion

The chlorophyll *a* bioassay indicates that algal biomass in 1988 was controlled by nutrients with little influence of grazers. Algal biomass on the disks increased with nutrient addition but did not increase any further with removal of grazers (Fig. 7; Table 3). The only significant treatment was the addition of nutrients (NP or NPM treatments); no other treatment affected algal biomass on the disks (Fig. 7; Table 3). Contrast analysis indicated that exclusion of grazers using malathion did not increase the chlorophyll response, although the addition of nutrients (NP or NPM) did increase the chlorophyll response relative to the appropriate control (Table 3). The algal response to added N and P in the P-fertilized reach may indicate a secondary limitation by N.

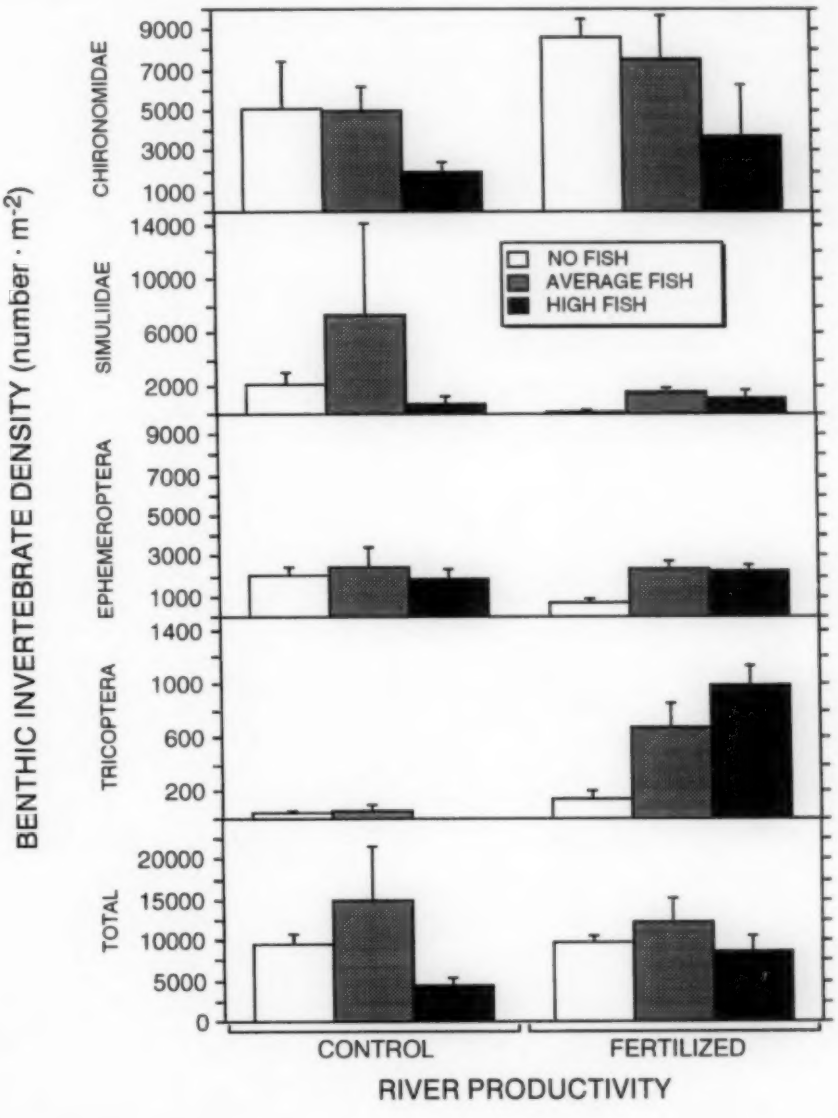
Response of Arctic grayling to fertilization and fish density

Fish growth was low in 1988 and strongly affected by fish density, although not by river fertilization (Fig. 8; Table 4). Fish weight change was lower in the high fish density treatments compared with the average fish density treatments (contrast analysis; weight: $F = 21.4$, $P = 0.0001$). Arctic grayling lost weight in the high fish abundance treatments (–8.9 g in the control and –19.9 g in the fertilized reach) and gained little weight (5.2 g in the control and 6.1 g in the fertilized reach) in the average fish abundance treatments (Fig. 8). Changes in total length were small and within measurement error for all treatments (Fig. 8). In the high fish density treatments, growth was lower in the fertilized compared with the control reach, possibly because of the influx of over 100 fish in the last 5–7 days (15%) of the experiment.

For both males and females, lipid storage was not different between fertilized and control reaches within fish density treatments (Fig. 9; Table 4). Both long-term (neutral lipid storage) and short-term (HSI) energy stores were lower in the high fish density treatment compared with the average fish density treatment (Fig. 9; Table 4). Contrast analysis indicated lower lipid storage in high fish density treatments for both females

© 1997 NRC Canada

Fig. 5. Abundance of benthic invertebrates (±1 SE) in river productivity (control or fertilized) and fish abundance (no fish, average fish, or high fish) treatments.



and males compared with the average fish density treatment (females: lipid, $F = 5.1$, $P = 0.01$; HSI, $F = 4.4$, $P = 0.04$; males: lipid, $F = 4.3$, $P = 0.0002$; HSI, $F = 9.8$, $P = 0.003$). In the fertilized reach, lipid storage in females was 70% lower and male lipid storage was 74% lower in the high fish density treatment compared with the average fish density treatment. In the control reach, lipid storage in females was 37% lower and lipid storage in males was 13% lower in the high fish density treatment compared with the average fish density treatment.

Gonad mass in both males and females was not affected by river fertilization; however, female gonad mass was affected by fish density (Fig. 9). Contrast analysis indicated that females in the average fish density treatments had more gonad mass than those in the high fish density treatments ($F = 14.45$, $P = 0.0007$). Males had an average gonad mass of about 3.4 g regardless of fertilization or fish density (Fig. 9).

Comparison of standing stocks among trophic levels

We scaled all measurements of standing stocks to a pool/riffle reach roughly 100 m long × 10 m wide to compare food availability among trophic levels for the high and average fish density treatments. The ratio of chlorophyll *a* to the number of benthic insects (1°:2°) increased from around 750 in the control reach to around 3000 in the fertilized reach because algal abundance increased, although total insect abundance remained roughly the same. The ratio of chlorophyll *a* per

© 1997 NRC Canada

Table 2. ANOVA results of invertebrate response to fish density – river fertilization treatments.

	Benthic assemblage[a]			Drift assemblage[b]							
	Treatment		Residual	Treatment		Time		Treatment–time		Subject (group)	Time–subject
	MS	*F*	MS	MS	*F*	MS	*F*	MS	*F*	MS	MS
Total	0.40	2.57	0.15	0.14	4.72	0.87	7.95	0.06	0.54	0.03	0.11
Simulidae	4.10	2.06	1.99	0.09	12.39*	0.23	5.94	0.02	0.50	0.01	0.04
Chironomids	1.18	2.0	0.58	0.08	1.45	0.74	15.12*	0.14	2.92	0.06	0.05
Tricoptera	20.15	9.38*	2.15	0.03	3.25	0.38	101.78*	0.02	4.48	0.01	0.01
Ephemoptera	0.65	3.33	0.19	0.24	8.08	0.44	13.33*	0.03	0.77	0.03	0.03

Note: *Significant at $P < 0.05$ level using Bonferroni adjustment ($n = 5$).

[a]Degrees of freedom: treatment, 5; residual, 12.

[b]Degrees of freedom: treatment, 5; time, 1; treatment·time, 5; subject (group), 6; time·subject (group), 6.

Fig. 6. Abundance of invertebrate drift (±1 SE) in river productivity (control or fertilized) and fish abundance (no fish, average fish, or high fish) treatments.

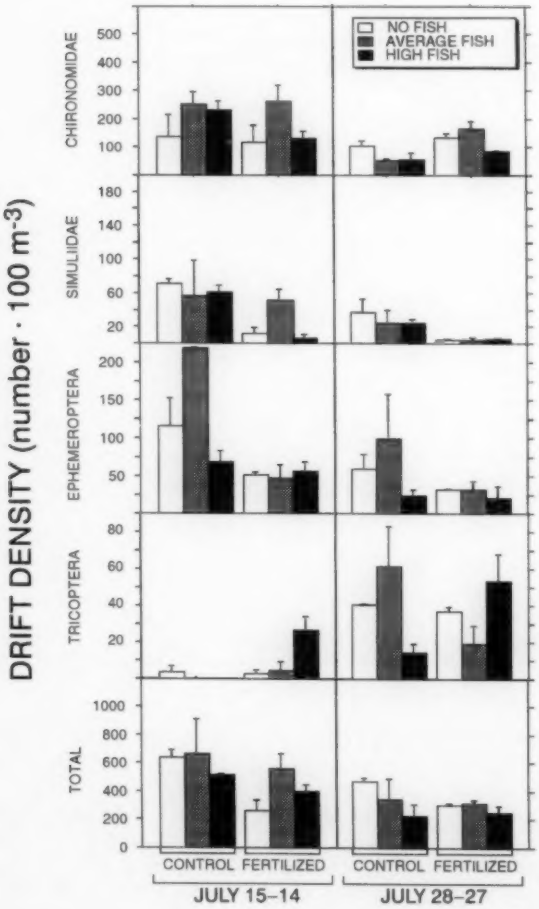
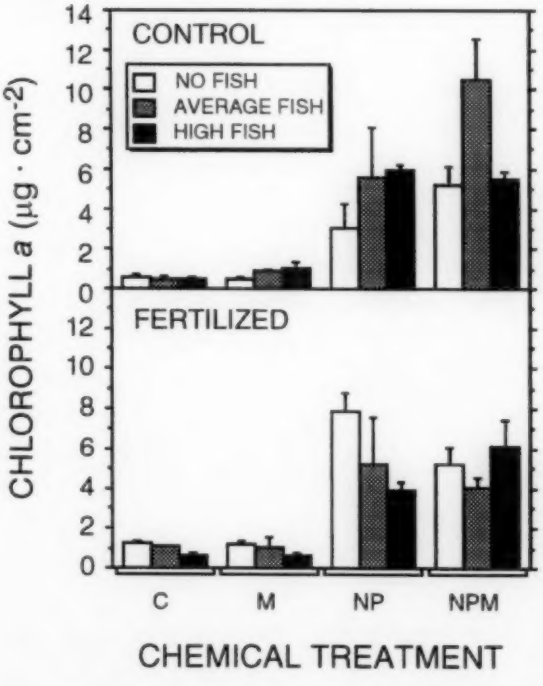


Fig. 7. Response of the epilithon bioassay (chlorophyll *a* per disk) (±1 SE) to river productivity, fish abundance, and chemical addition. River productivity was control or fertilized; fish density was no fish, average fish, or high fish density; chemical treatment was no treatment (C), malathion (M), nitrogen + phosphorus (NP), and nitrogen + phosphorus + malathion (NPM).



drifting insect was less variable, ranging from 18 to 26 across all treatments. The ratio of total benthic insects per fish (2°:3°) decreased over an order of magnitude between the average (107×10^4 to 154×10^4 benthic insects per fish) and high (4×10^4 to 7×10^4 benthic insects per fish) fish density treatment. The ratio of total daily drifting insects to fish also decreased an order of magnitude between the average (1270–2731 drifting insects per fish) and high (115–160 drifting insects per fish) fish density treatments. This indicated that

© 1997 NRC Canada

Table 3. ANOVA results of chlorophyll *a* bioassay response to fish density – river productivity treatment and chemical addition.

	df	MS	*F*
ANOVA factor			
Treatment	5	0.22	1.72
Chemical	3	9.09	70.20*
Treatment–chemical	15	0.15	1.12
Residual	47	0.13	
Contrast analysis			
Test: insect grazing effect			
C = M			0.38
NP = NPM			3.02
Test: nutrient effect, no additional grazing			
C = NP			94.35*
C, M = NP, NPM			206.51*

Note: *Significant at $P < 0.05$ level.

Table 4. ANOVA results of fish response to fish density and river fertilization.

	Treatment		Residual
	MS	*F*	MS
Change in:[a]			
weight	2568.94	7.71*	333.13
length	0.80	7.39*	0.11
condition factor	0.01	2.7	0.01
Neutral lipid[b]			
Female	1.92	2.50	0.76
Male	2.93	11.74*	0.25
HSI[b]			
Female	0.05	2.11	0.02
Male	0.08	4.14*	0.02
Gonad[b]			
Female	37.83	6.04*	6.26
Male	0.76	0.67	1.12

Note: *Significant at the $P < 0.05$ level.

[a]Degrees of freedom for *F* tests: treatment, 3; residual, 76.

[b]Degrees of freedom for *F* tests: treatment, 3; residual: female lipid, 29; male lipid, 39; female gonad, 31; male gonad, 40; female HSI, 30; male HSI, 39.

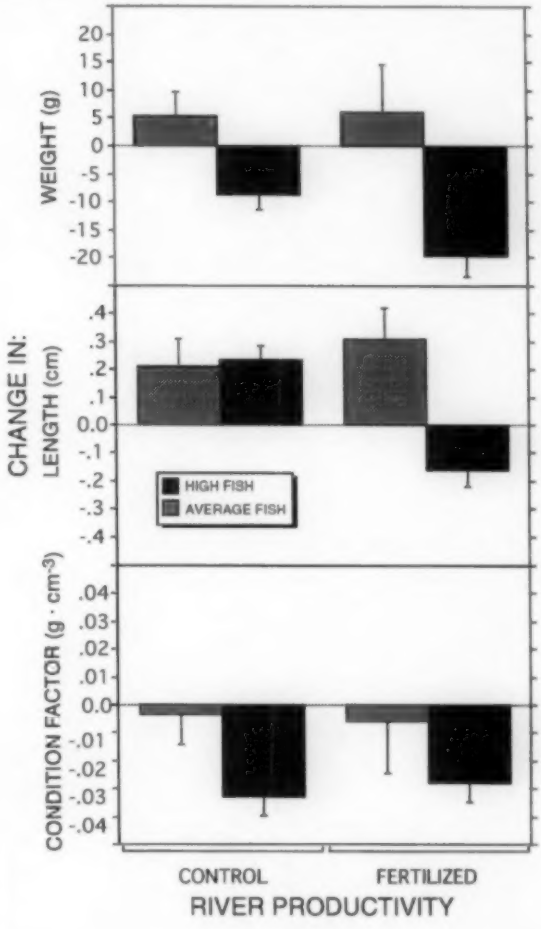
fish growth was more highly correlated with per capita insect availability than with per capita algal standing stock (Table 5).

Discussion

Overview

The results of the whole-river fertilization and fish density experiments in 1988 contrast with earlier years (1985–1987) when insect grazer control of algal biomass was observed (Gibeau and Miller 1989; Peterson et al. 1993*a*). In 1988, algal biomass accumulation in the fertilized reach was higher than that seen in the first 2 years of experimental fertilization (1983–1984) when insects did not exert strong grazer control.

Fig. 8. Average change (±1 SE) in fish weight, length, and condition factor in river productivity (control or fertilized) and fish density (average fish or high fish) treatments.

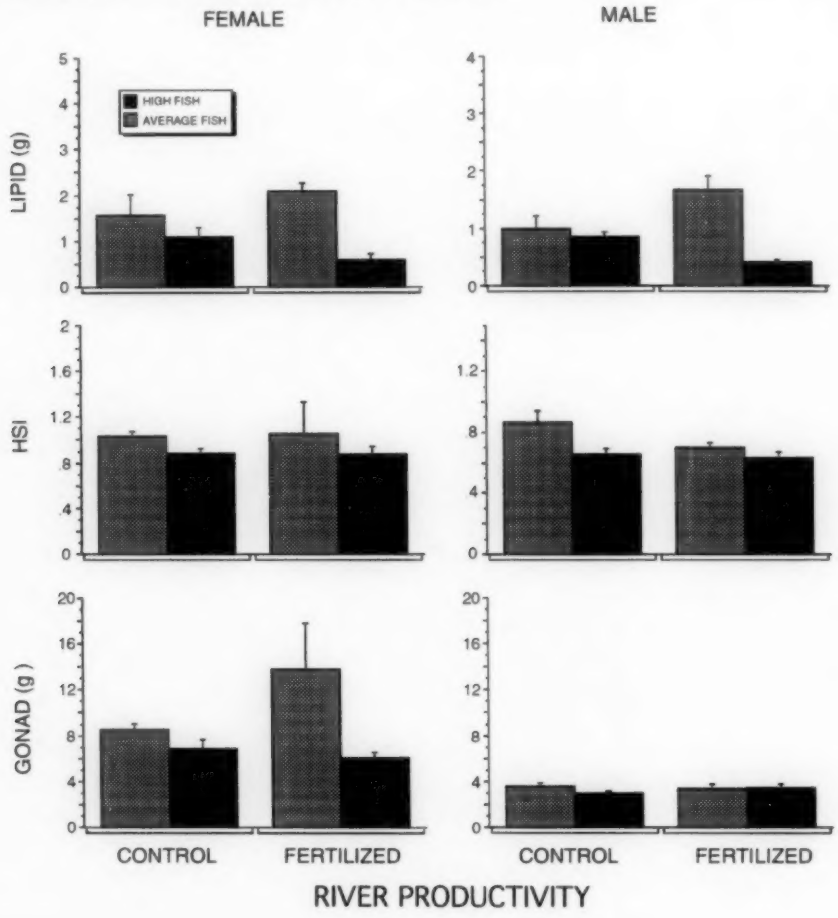


A grazer bioassay using malathion treatments also indicated little effect of insect grazers on algal biomass in 1988. If insect grazers were strongly controlling algal biomass, we expected an increase in chlorophyll *a* on malathion-treated disks compared with control disks (C versus NP versus NPM). If top-down control by fish occurred, we expected algal biomass to have increased in the high fish density treatment because insect grazing on algae should have decreased. In 1988, increasing and decreasing predation pressure on insect grazers by manipulation of fish density did not result in either large declines in insect densities or increases in algal biomass (Table 5). Fish growth was, however, strongly influenced by per capita availability of food, suggesting that their production is limited by competition for food.

One difficulty with the interpretation of the results of this experiment is the lack of explicit treatment replication (Stewart-Oaten et al. 1992). Each treatment in our study covers a very large spatial scale (treatment length of 100–500 m). Thus, we avoid alterations of substrate resulting from trapping of sediment or detritus by cages and small-scale sampling

© 1997 NRC Canada

Fig. 9. Neutral lipid storage, hepatosomatic index (HSI), and reproductive mass (gonad) (±1 SE) for male and female Arctic grayling in river productivity (control or fertilized) and fish density (average or high fish) treatments.



effects common to other studies (Cooper et al. 1990; Peckarsky and Penton 1990; Lancaster et al. 1991; Downes et al. 1993; Dudgeon 1993). We were, however, unable to replicate the treatments because of the distance of fertilized stream available and the number of Arctic grayling needed in each treatment to evaluate the response of the top consumer. Even with this lack of explicit replication, we believe that this experiment offers insight into factors regulating stream production and top consumers. One form of replication planned for the future is to examine the effects of similar treatments in a second river.

Grazer control of algal standing stock

The lack of grazer control of the algal standing stock may have been due to several factors: the exceptionally good conditions for algal growth, a change in algal species to less edible forms, and a change in the insect grazing pressure. Midsummer spates contribute to the control of epilithic algae, in some cases scouring up to 50% of the epilithic biomass (Hullar and Vestal 1989). In 1988, we had low flow until late in the season, which provided good conditions for algal growth. As a result, long strands of filamentous algae that are less susceptible to grazing by mayflies than are epilithic diatoms developed.

Another explanation for the accumulation of high algal biomass is a change in insect grazing pressure as a result of changes in insect community composition or abundance. Mayflies, a dominant grazer, have increased in abundance since the first year of fertilization. Benthic mayfly densities were roughly 100–400·m^{-2} in the first year and increased to 1000 to over 6000·m^{-2} in subsequent years (Peterson et al. 1993*a*). In 1988, mayfly densities were 3000·m^{-2}, in about the middle of the range of abundance. Other insects, such as small chironomids, may also be important grazers in streams (Power 1990). Chironomids were not very abundant in 1988 (5000·m^{-2}). The moderate abundance of all grazing insects combined with physical conditions conducive to good algal growth may have released the algae from top-down control by grazers.

© 1997 NRC Canada

Table 5. Abundance (normalized to one 100 × 10 m pool\riffle reach) and ratios of primary, secondary, and tertiary producers in the average and high fish density treatments in control and fertilized river reaches.

		Control		Fertilized	
		Average	High	Average	High
Primary (1°)	Chl *a* (μg × 10^6)[a]	1500	1500	9000	9000
Secondary (2°)	Benthic invertebrates (no. × 10^4)[b]	448	135	366	256
	Total drift invertebrates (no. · 24 h^{-1})[c]	7920	5760	4320	4320
	Mayflies and caddisflies[d,c]	3960	2880	2160	2160
Tertiary (3°)	Fish density (no.)	2.9	35.8	3.4	37.5
	Per capita fish growth (g weight)	6	−7	+8	−20
	(g lipid)	1.2	1	1.7	0.5
1°:2°	Chl *a*/benthic	334	1111	2459	3515
	Chl *a*/drift (×10^4)	18	26	20	20
1°:3°	Chl *a*/fish (×10^6)	517	41	2647	240
2°:3°	Benthic/fish density (×10^4)	154	4	107	7
	Total drift/fish density	2731	160	1270	115
	Mayflies and caddisflies	1365	80	635	58

[a]Chlorophyll (control, 5 × 10^6 μg · m^{-2}; fertilized, 30 × 10^6 μg · m^{-2}) × 300 m^2 of riffle (30 m long × 10 m wide).

[b]Benthic insects: standing stocks from each treatment (control:average = 14 935, high = 4526; fertilized: average = 12 230, high = 8527 · m^{-2}) × 300 m^2 of riffle.

[c]Drift: average number of individuals · 100 m^{-3} by treatment (control: average = 550, high = 400; fertilized: average = 300, high = 300) × 14.4 100 m^3 · 24 h^{-1}.

[d]Mayflies and caddisflies; the preferred food of grayling are roughly 40–50% of the total drift.

Response of fish to whole-stream enrichment and fish density changes

Adult Arctic grayling production in the Kuparuk River is clearly influenced by the per capita availability of food resources. In both the control and fertilized reaches of the river, fish growth and energy storage declined with increased fish density. Higher growth in the fertilized reach compared with the control reach, although not great in 1988, has been consistent throughout the years of fertilization (Deegan and Peterson 1992). The density-dependent growth and response to fertilization in most years both indicate that Arctic grayling are food limited.

Interestingly, the difference in adult Arctic grayling growth between the high and average fish abundance treatments was not as great as expected. A 10-fold increase in fish density resulted in only a twofold decrease (from 6 to −10 g) in fish growth. The relatively small differences in growth between fish density treatments may have been related to the overall poor conditions for growth in 1988. The small weight gains of fish in the average-density treatments and the ratio of insects drifting per fish (Table 5) give some indication of the minimum insect drift production required for growth (Table 5). The high fish treatment densities were too high relative to the availability of insects to satisfy the minimum maintenance requirements of the fish, as indicated by weight loss over the course of the experiment.

Salmonid growth is often thought to be limited by a combination of food availability and fish density. Several surveys have found positive correlation between nutrient concentrations and growth of fish under similar fish densities, indicating that growth was limited by per capita food availability (McFadden and Cooper 1962; Egglishaw 1968; Murphy et al. 1981; Wilzbach 1985; Bowlby and Roff 1986; Gibson and Haedrich 1988). McFadden and Cooper (1962) found that streams carrying higher concentrations of dissolved solids supported a greater standing crop of brown trout (*Salmo trutta*) than those with lower "fertility." Salmonid biomass at five stream sites in Newfoundland was apparently related to the type of habitat, stability of discharge, and level of nutrients (Gibson and Haedrich 1988). In addition, several experimental field studies support these observations. Warren et al. (1964) observed that a fourfold increase in invertebrate production, induced by adding sucrose to a stream, resulted in a sevenfold increase in the weight gain of YOY trout in a hatchery stream. Mason (1976) increased the growth of YOY salmonids by supplemental feeding in a natural stream. Johnston et al. (1990) demonstrated that N and phosphate addition increased growth of young salmonids, presumably by stimulating algae and insect production.

Lower food per capita (Table 5) coupled with increased energy demands for aggressive behavior possibly resulted in weight loss in the high fish density treatments. At high adult fish densities, the energy costs of aggressive behavior to defend territories may also become significant (Wooton 1985; Hughes and Dill 1990; Hughes 1992). Weight loss was most pronounced in the high fish abundance treatment in the fertilized reach, probably because of the influx of at least 100 individuals late in the experiment. After this influx (the last 5–7 days, 15% of the growing season), we saw fish chasing and fighting each other. These types of behaviors were not observed in the average fish abundance treatments.

Environmental conditions also strongly affected adult

© 1997 NRC Canada

Arctic graying growth. Fertilization increased Arctic grayling growth most in years of moderate river flow and temperature. Growth of fish at average density in 1988 (5–6 g) was quite low compared with the 4-year average (control: 24 g; fertilized: 55 g) and was similar to that in 1990 (control: 2 g; fertilized: 6 g), another year of low flows and high temperatures. We observed little to no growth of adult fish in 2 years (1988 and 1990) with low flows and high temperatures (Deegan and Peterson 1992). Low stream flows and high temperatures affect the energy balance of Arctic grayling by decreasing insect drift rates and increasing Arctic grayling energy demands (Tytler and Calow 1985). Insect drift rates are lower in areas with higher insect food availability (Kohler 1985; Hinterleitner-Anderson et al. 1992) and during low stream flows (LaPerriere 1983). In 1988, the density of insects drifting in the fertilized reach may have been low enough that fish stopped feeding or at least had trouble locating the sparse drifting insects. Reduced feeding activities resulting from the unusually high temperatures during this study could also have contributed to low growth, as has been suggested for brook trout (*Salvelinus fontinalis*) (Bechara et al. 1992).

Top-down role of fish

The role of Arctic grayling as an effective top-down control in the Kuparuk River is difficult to determine based on this experiment. Because Arctic grayling are primarily drift feeders, we expected a greater impact on drift compared with benthic insect abundances. We also expected, based on selective feeding by Arctic grayling, that mayflies and caddisflies would be the most affected and chironomids and blackflies the least affected by fish predation. Comparison of the energy demands of Arctic grayling with the number of drifting invertebrates over 24 h indicates that Arctic grayling could consume a large fraction of the available insect drift. Using the Wisconsin bioenergetics model (Hewett and Johnson 1992) calibrated for Arctic grayling and the temperature and growth of the summer of 1988, we estimated that a single adult Arctic grayling consumed about 1500 mayflies or 240 caddisflies in 24 h. At average fish densities in the control reach, roughly 1365 mayflies and caddisflies per fish passed through a pool in a 24-h period. Most of these insects are concentrated in preferred feeding lanes of Arctic grayling and should be vulnerable to predation (Hughes 1992; O'Brien and Schowalter 1993). Although this comparison is rough, it indicates that Arctic grayling should have an impact on the drift community. It also indicates that the drift per individual fish (58–80) in the high-fish treatments could not possibly supply sufficient food for growth. However, we detected no difference in drift or insect abundance associated with fish density.

The effect of fish predation on insect abundance was difficult to determine in part because of some of the assumptions of our sampling design and in part because of the nature of aquatic insect populations. The high small-scale variability of insect abundances (Downes et al. 1993), colonization from upstream sources (Cooper et al. 1990), and the behavioral response of insects to food availability and predators (Sih and Wooster 1994) all combine to make assessing the impacts of predators difficult. Downes et al. (1993) have shown that benthic faunal abundances can vary considerably over scales from stones within riffles to sites within reaches. Because we had to assume that insect abundances were relatively uniform within productivity reaches before fish density treatments, and variation among riffles within a treatment was higher than anticipated, our probability of detecting differences among treatments was low. We are not sure that this difficulty can be overcome, however, because insect communities also change over time because of drift and emergence, thus ruling out any "before and after" types of analyses.

Colonization from upstream sources could have also masked the effects of fish predation on insects. Cooper et al. (1990) showed that when drift is abundant, high colonization rates tend to compensate for the effects of predation. They found a significant impact of fish when drift rates, and hence recruitment, were low (3–5 mayfly nymphs·100 m^{-3}), although at higher drift densities (50–1300 nymphs·100 m^{-3}), fish predation effects were not detected. Our mayfly drift densities were near the lower end of this high range, indicating that replenishment from upstream could have masked the effects of fish predation. This problem was exacerbated by our placement of treatments because increased drift into a "high-fish" treatment from a "no-fish" treatment could have contributed to masking the effect of fish predation. Although this arrangement of treatments provided a conservative test of our hypothesis, it also might obscure a real effect. If we were to redo this experiment, we would sample both immediately upstream and downstream of fish treatments to differentiate the effects of recruitment from "no-fish impact."

There may also be a relationship between the stream productivity and the importance of fish predation in influencing ecosystem dynamics. Whether top-down control occurs is also a function of predator efficiency (Power 1992*a*; Abrams 1993). Fertilization may alter these relationships because increased food availability may result in decreased foraging or drift by the insects, thus changing their susceptibility to predation (Abrams 1993). In addition, insects may respond to the presence of predators by either hiding under rocks or dispersing and may change their behavioral response to predators if food availability changes (Sih and Wooster 1994).

Some studies have suggested that trophic cascades are most probable in oligotrophic, low-productivity systems because grazers consume a larger fraction of the primary production (Feminella and Hawkins 1995). The probability of top-down control of insects by Arctic grayling may be higher in the low-productivity (control) reach where the food requirements of Arctic grayling are high relative to insect abundance and insects are more vulnerable to predation because they drift more often in response to low algal availability. Control of grazing insects by Arctic grayling in the low-productivity reach would probably not result in a trophic cascade, however, because the scope of the algal response in the low-productivity reach is controlled by nutrient availability. Grazer removal bioassays indicate a limited response to insect grazer removal in the control reach (M.C. Miller, personal observation).

In addition, fish predation also may not cause a trophic cascade if the fish are eating insects as they are emerging, in essence after the insects have ceased grazing on algae. Adult Arctic grayling prey heavily and preferentially on larger sized nymphs of mayflies and caddisflies and do not take small chironomids in proportion to their availability in the drift. Early in the summer, when small mayfly nymphs are drifting in search of food, Arctic grayling tend to take the larger, emerging caddisflies. Caddisflies are primarily filter feeders, not

© 1997 NRC Canada

grazers, and do not directly affect epilithic algal abundance. Later in the summer, Arctic grayling feed heavily on mayflies, both larger drifting nymphs and emerging adults. Adult Arctic grayling rarely feed on small drifting nymphs or emerging adult chironomids. The influence of Arctic grayling predation on the accumulation of algal biomass will depend on the relative abundance and use of these different food sources by Arctic grayling and the relative strength of the grazing pressure of each of these insect groups.

Another possibly important interaction in the Kuparuk River is the role that YOY Arctic grayling may have on the insect community. YOY abundance is highly variable (L.A. Deegan, personal observation). In the early years (1983–1987) of the experiment, YOY abundances were sufficiently low that we did not feel that they could exert any regulatory pressure on insects. In 1988, however, YOY abundance and end-of-year size reached a record high throughout all reaches of the river (Deegan and Peterson 1992). YOY Arctic grayling also feed on insects, specializing in small chironomids and early instars of *Baetis* when the fish are small and gradually eat larger items as they grow (L.A. Deegan and H. Golden, personal observation). The lack of insect control of algae in all reaches of the river during 1988 could have been due to the record number of YOY Arctic grayling that consumed small insects and instars that adult Arctic grayling do not eat. We are currently doing caging experiments with several densities of YOY to test this possibility (L.A. Deegan and H. Golden, personal observation).

There is currently no consensus on the importance of fish in structuring stream ecosystem dynamics across many temperate, tropical, and now arctic streams. This is surprising in view of "Allen's paradox" that insect production in streams is often too low to support observed fish densities. Only studies that examine the effects of algivorous fish in streams find consistent effects on ecosystem dynamics (Matthews et al. 1987; Gelwick and Matthews 1992; Power 1992*b*; Vaughn et al. 1993). Trophic cascades are more difficult to demonstrate. Some researchers who have experimentally examined fish predation in the field have shown that although fish feed heavily on stream insects, they play a minor role in determining insect abundance and diversity (Allan 1982; Flecker and Allan 1984; Culp 1986; Reice and Edwards 1986). Others have found that fish predation primarily affects larger, more motile insects, particularly in pools (Hemphill and Cooper 1984; Wilzbach 1985; Schlosser and Ebel 1989; Dudgeon 1993; Wiseman et al. 1993). Griffiths (1981) increased the density of trout by three- to four-fold over normal densities and found that some insect species increased in abundance and others decreased. Several studies have demonstrated that fish predation effects vary with river flow and habitat complexity (Gilliam et al. 1989; Schlosser and Ebel 1989; Bechara et al. 1992; Holomuzki and Stevenson 1992; Dudgeon 1993). There probably is no universal role of fish in structuring stream ecosystems, but rather their role needs to be considered in the context of other ecosystem factors (such as light, organic matter sources, and disturbances), as well as foraging interactions and behavioral responses.

There are clearly both bottom-up and top-down forces operating in stream ecosystems. In the Kuparuk River, the control of benthic chlorophyll levels by grazing insects has proven to be variable from year to year. This experiment was performed in a low-flow year when grazers had little effect on algae and fish had little effect on grazers. We conclude that the strength of the biotic interaction is variable from year to year as the result of large variations in physical factors (10-fold mean discharge variation) and to large variations (10-fold or greater) in abundance of algae, grazers, and fish, suggesting that the probability of any one experiment demonstrating the strength of several interactions simultaneously may be quite low.

Acknowledgments

Many of the ideas presented in this paper are the result of stimulating and sometimes heated debate with everyone on the Kuparuk River fertilization research team. We thank them all for their help and criticisms. C. Bauman, T. Duncan, and C. Neill deserve special thanks for field work. This work was funded by National Science Foundation grants (OPP-9024188, DEB-9211775, and DEB-870232), the National Geographic Society, and the W. Mellon Foundation.

References

Abrams, P.A. 1993. Effect of increased productivity on the abundances of trophic level. Am. Nat. **141**: 351–371.

Allan, J.D. 1982. The effect of reduction of trout density on the invertebrate community of a mountain stream. Ecology, **63**: 1444–1455.

Bechara, J.A., Moreau, G., and Planas, D. 1992. Top-down effects of brook trout (*Salvelinus fontinalis*) in a boreal forest stream. Can. J. Fish. Aquat. Sci. **49**: 2093–2103.

Bolger, T., and Connolly, P.L. 1989. The selection of suitable indices for the measurement and analysis of fish condition. J. Fish Biol. **34**: 171–182.

Bowlby, J.N., and Roff, J.C. 1986. Trout biomass and habitat relationships in southern Ontario streams. Trans. Am. Fish. Soc. **115**: 503–514.

Carpenter, S.R. 1990. Large scale perturbations: opportunities for innovation. Ecology, **71**: 2038–2043.

Carpenter, S.R., Kitchell, J.F., Hodgson, R., Cochran, P.A., Elser, J.J., Lodge, D.M., Kretchmer, D., He, X., and Von Ende, C.N. 1987. Regulation of lake primary productivity by food web structure. Ecology, **68**: 1863–1876.

Carpenter, S.R., Frost, T.M., Kitchell, J.F., Kratz, T.K., Schindler, D.W., Shearer, J., Sprules, W.G., Vanni, M.J., and Zimmerman, A.P. 1990. Patterns of primary production and herbivory in 25 North American lake ecosystems. *In* Comparative analyses of ecosystems: patterns, mechanisms and theories. *Edited by* J. Cole, S. Findlay, and G. Lovett. Springer-Verlag, New York. pp. 67–96.

Cooper, S.D., Walde, S.J., and Peckarsky, B.L. 1990. Prey exchange rates and the impact of predators on prey populations in streams. Ecology, **71**: 1503–1514.

Culp, J.M. 1986. Experimental evidence that stream macroinvertebrate community structure is unaffected by different densities of coho salmon fry. J. N. Am. Benthol. Soc. **5**: 140–149.

Deegan, L.A., and Peterson, B.J. 1992. Whole river fertilization stimulates fish production in an arctic tundra river. Can. J. Fish. Aquat. Sci. **49**: 1890–1901.

Downes, B.J., Lake, P.S., and Schreiber, E.S.G. 1993. Spatial variation in the distribution of stream invertebrates: implications of patchiness for models of community organization. Freshwater Biol. **30**: 119–132.

© 1997 NRC Canada

Dudgeon, D. 1993. The effects of spate-induced disturbance, predation and environmental complexity on macroinvertebrates in a tropical stream. Freshwater Biol. **30**: 189–197.

Eberhardt, L.L., and Thomas, J.M. 1991. Designing environmental field studies. Ecol. Monogr. **61**: 53–73.

Egglishaw, H.J. 1968. The quantitative relationship between bottom fauna and plant detritus in streams of different calcium concentrations. J. Appl. Ecol. **5**: 731–740.

Elwood, J.N., Newbold, J.D., Trimble, A.F., and Stark, R.W. 1981. The limiting role of phosphorus in a woodland stream ecosystem: effects of P enrichment on leaf decomposition and primary producers. Ecology, **62**: 146–158.

Feminella, J.W., and Hawkins, C.P. 1995. Interactions between stream herbivores and periphyton: a quantitative analysis of past experiments. J. N. Am. Benthol. Soc. **14**: 465–509.

Flecker, A.S., and Allan, J.D. 1984. The importance of predation substrate and spatial refuge in determining lotic insect distributions. Oecologia (Berl.), **64**: 305–313.

Fretwell, S.D. 1987. Food chain dynamics: the central theory of ecology? Oikos, **50**: 291–301.

Gardiner, W.R., and Geddes, P. 1980. The influence of body composition on the survival of juvenile salmon. Hydrobiologia, **69**: 67–72.

Gelwick, F.P., and Matthews, W.J. 1992. Effects of an algivorous minnow on temperate stream ecosystem properties. Ecology, **73**: 1630–1645.

Gibeau, G.G., and Miller, M.C. 1989. A micro-bioassay for epilithon using nutrient diffusing artificial substrata. J. Freshwater Ecol. **5**: 171–176.

Gibson, R.J., and Haedrich, R.L. 1988. The exceptional growth of juvenile Atlantic salmon (*Salmo salar*) in the city waters of St. Johns, Newfoundland, Canada. Pol. Arch. Hydrobiol. **35**(3/4): 385–407.

Gilliam, J.F., Fraser, D.F., and Sabat, A.M. 1989. Strong effects of foraging minnows on a stream invertebrate benthic community. Ecology, **70**: 445–452.

Griffiths, R.W. 1981. The effect of trout predation on the abundance and production of stream insects. Masters thesis, University of British Columbia, Vancouver, B.C.

Hemphill, N., and Cooper, S.D. 1984. Differences in the community structure of stream pools containing or lacking fish. Verh. Int. Ver. Limnol. **22**: 1858–1861.

Hershey, A.E., and Hiltner, A.E. 1988. Effects of caddisfly activity on black fly density: interspecific interactions outweigh food limitation. J. N. Am. Benthol. Soc. **7**: 188–196.

Hershey, A.E., Hiltner, A.E., Hullar, M.A.J., Miller, M.C., Vestal, J.R., Lock, M.A., Rundle, S., and Peterson, B.J. 1988. Nutrient influence on a stream grazer: *Orthocladius* microcommunities respond to nutrient input. Ecology, **69**: 1383–1392.

Hewett, S., and Johnson, B. 1992. Fish bioenergetics model 2. A generalized bioenergetics model of fish growth for microcomputers. University of Wisconsin Sea Grant Institute, Madison, Wis.

Hiltner, A.L., and Hershey, A.E. 1992. Blackfly (Diptera: Simuliidae) responses to phosphorus enrichment of an arctic tundra stream. Hydrobiologia, **240**: 259–265.

Hinterleitner-Anderson, D., Hershey, A.E., and Schuldt, J.A. 1992. The effects of river fertilization on mayfly (*Baetis* sp.) drift patterns and population density in an arctic river. Hydrobiologia, **240**: 247–258.

Holomuzki, J.R., and Stevenson, R.J. 1992. Role of predatory fish in community dynamics of an ephemeral stream. Can. J. Fish. Aquat. Sci. **49**: 2322–2330.

Huang, C., and Sih, A. 1990. Experimental studies on behaviorally mediated, indirect interactions through a shared predator. Ecology, **71**: 1515–1522.

Hughes, N.F. 1992. Ranking of feeding positions by drift-feeding arctic grayling (*Thymallus arcticus*) in dominance hierarchies. Can. J. Fish. Aquat. Sci. **49**: 1994–1998.

Hughes, N.F., and Dill, L.M. 1990. Position choice by drift-feeding salmonids: model and test for arctic grayling (*Thymallus arcticus*) in subarctic mountain streams, interior Alaska. Can. J. Fish. Aquat. Sci. **47**: 2039–2048.

Hullar, M.A., and Vestal, J.R. 1989. The effects of nutrient limitation and stream discharge on the epilithic microbial community in an arctic stream. Hydrobiologia, **172**: 19–26.

Hurlbert, S.H. 1984. Pseudoreplication and the design of ecological field experiments. Ecol. Monogr. **54**: 187–211.

Johnson, C.R., and Field, C.A. 1993. Using fixed-effects model multivariate analysis of variance in marine biology and ecology. Oceanogr. Mar. Biol. Annu. Rev. **31**: 177–221.

Johnston, N.T., Perrin, C.J., Slaney, P.A., and Ward, B.R. 1990. Increased juvenile salmonid growth by whole-river fertilization. Can. J. Fish. Aquat. Sci. **47**: 862–872.

Kohler, S.L. 1985. Identification of stream drift mechanisms: an experimental and observational approach. Ecology, **66**: 1749–1761.

Kriet, K., Peterson, B.J., and Corliss, T.L. 1992. Water and sediment export of the Upper Kuparuk River drainage of the North Slope of Alaska. Hydrobiologia, **240**: 71–81.

Lancaster, J., Hildrew, A.J., and Townsend, C.R. 1991. Invertebrate predation on patchy and mobile prey in streams. J. Anim. Ecol. **60**: 625–641.

LaPerriere, J.D. 1983. Alkalinity, discharge, average velocity, and invertebrate drift concentration in subarctic Alaskan streams. Freshwater Ecol. **2**: 141–151.

Lock, M.A., Ford, T.E., Fiebig, D.M., Miller, M.C., Hullar, M., Vestal, J.R., and Peterson, B.J. 1989. A biogeochemical survey of rivers and streams in the mountains and foothills province of arctic Alaska. Hydrobiology, **115**: 499–521.

Mason, J.C. 1976. Response of underyearling coho salmon to supplemental feeding in a natural stream. J. Wildl. Manage. **40**: 775–788.

Matthews, W.J., Stewart, A.J., and Power, M.E. 1987. Grazing fishes as components of North American stream ecosystems: effects of *Campostoma anomalum*. *In* Community and evolutionary ecology of North American stream fishes. *Edited by* W.J. Matthews and D.C. Heins. University of Oklahoma Press, Norman, Okl. pp. 128–135.

McFadden, J.T., and Cooper, E.L. 1962. An ecological comparison of six populations of brown trout (*Salmo trutta*). Trans. Am. Fish. Soc. **91**: 53–62.

Murphy, M.L., Hawkins, C.P., and Anderson, N.H. 1981. Effects of canopy modification and accumulated sediment on stream communities. Trans. Am. Fish. Soc. **110**: 469–478.

O'Brien, W.J., and Schowalter, J.J. 1993. Effects of current velocity and suspended debris on the drift feeding of Arctic grayling. Trans. Am. Fish. Soc. **122**: 609–615.

Ohman, M.D. 1988. Sources of variability in measurements of copepod lipids and gut fluorescence in the California Current coastal zone. Mar. Ecol. Prog. Ser. **42**: 143–153.

Oksanen, L., Fretwell, S.D., Arruda, J., and Niemela, P. 1981. Exploitation ecosystems in gradients of primary productivity. Am. Nat. **118**: 240–261.

Oksanen, T. 1990. Exploitative ecosystems in heterogeneous habitat complexes. Evol. Ecol. **4**: 220–234.

Peckarsky, B.L., and Penton, M.A. 1990. Effects of enclosures on stream microhabitat and invertebrate community structure. J. N. Am. Benthol. Soc. **9**: 249–261.

Persson, L., Diehl, S., Johansson, L., Andersson, G., and Hamrin, S.F. 1992. Trophic interactions in temperate lake ecosystems: a test of food chain theory. Am. Nat. **140**: 59–84.

© 1997 NRC Canada

Peterman, R.M. 1990. Statistical power analysis can improve fisheries research and management. Can. J. Fish. Aquat. Sci. **47**: 2–15.

Peterson, B.J., Hobbie, J.E., Corliss, T.L., and Kriet, K. 1983. A continuous-flow periphyton bioassay: tests of nutrient limitation in a tundra stream. Limnol. Oceanogr. **28**: 583–591.

Peterson, B.J., Hobbie, J.E., Hershey, A.E., Lock, M.A., Ford, T.E., Vestal, J.R., McKinley, V.L., Hullar, M.A.J., Miller, M.C., Ventullo, R.M., and Volk, G.S. 1985. Transformation of a tundra river from heterotrophy to autotrophy by addition of phosphorus. Science (Wash., D.C.), **229**: 1383–1386.

Peterson, B.J., Hobbie, J.E., and Corliss, T.L. 1986. Carbon flow in tundra stream ecosystem. Can. J. Fish. Aquat. Sci. **43**: 1259–1270.

Peterson, B.J., Deegan, L., Helfrich, J., Hobbie, J.E., Hullar, M., Moller, B., Ford, T.E., Hershey, A., Hiltner, A., Kipphut, G., Lock, M.A., Fiebig, D.M., McKinley, V., Miller, M.C., Vestal, J.R., Ventullo, R., and Volk, G. 1993*a*. Biological responses of a tundra river to fertilization. Ecology, **74**: 653–672.

Peterson, B.J., Fry, B., Deegan, L., and Hershey, A. 1993*b*. The trophic significance of epilithic algal production in a fertilized tundra river ecosystem. Am. Soc. Limnol. Oceanogr. **38**: 872–878.

Power, M.E. 1990. Effects of fish in river food webs. Science (Wash., D.C.), **250**: 411–415.

Power, M.E. 1992*a*. Top-down and bottom-up forces in food webs: do plants have primacy? Ecology, **73**: 733–746.

Power, M.E. 1992*b*. Habitat heterogeneity and the functional significance of fish in river food webs. Ecology, **73**: 1675–1688.

Reed, R.J. 1964. Life history and migration patterns of Arctic grayling, *Thymallus arcticus* (Pallas), in the Tanana River drainage of Alaska. Research Report 2, Alaska Department of Fish and Game, Juneau, Alaska.

Reice, S.R., and Edwards, R.L. 1986. The effect of vertebrate predation on lotic macroinvertebrate communities in Quebec, Canada. Can. J. Zool. **64**: 1930–1936.

Rice, W.R. 1988. Analyzing tables of statistical tests. Evolution, **43**: 223–225.

Rosemond, A.D., Mulholland, P.J., and Elwood, J.W. 1993. Top-down and bottom-up control of stream periphyton: effects of nutrients and herbivores. Ecology, **74**: 1264–1280.

Schlosser, I.J., and Ebel, K.K. 1989. Effects of flow regime and cyprinid predation on a headwater stream. Ecol. Monogr. **59**: 41–57.

Sih, A., and Wooster, E.E. 1994. Prey behavior, prey dispersal, and predator impacts on stream prey. Ecology, **75**: 1199–1207.

Stewart, A.J. 1987. Responses of stream algae to grazing minnows and nutrients: a field test of interactions. Oecologia (Berl.), **72**: 1–7.

Stewart-Oaten, A., Bence, J.R., and Osenberg, C.W. 1992. Assessing the effects of unreplicated perturbations: no simple solutions. Ecology, **73**: 1396–1404.

Stockner, J.G., and Shortreed, K.R.S. 1978. Enhancement of autotrophic production by nutrient addition in a coastal rainforest stream on Vancouver Island. J. Fish. Res. Board Can. **35**: 28–34.

Strickland, J.D.H., and Parsons, T.R. 1972. A practical handbook of seawater analysis. Bull. Fish. Res. Board Can. No. 167.

Toft, C.A., and Shea, P.J. 1983. Detecting community-wide patterns: estimating power strengthens statistical inference. Am. Nat. **122**: 618–625.

Tytler, P., and Calow, P. 1985. Fish energetics: new perspective. Johns Hopkins University Press, Baltimore, Md.

Vaughn, C.C., Gelwick, F.P., and Matthews, W.J. 1993. Effects of algivorous minnows on production of grazing stream invertebrates. Oikos, **66**: 119–128.

Warren, C.E., Wales, J., Davis, G., and Doudoroff, P. 1964. Trout production in an experimental stream enriched with sucrose. J. Wildl. Manage. **28**: 617–660.

Wetzel, R.G., and Likens, G.E. 1979. Limnological analyses. W.B. Baunders, Philadelphia, Pa.

Wilzbach, M.A. 1985. Relative roles of food abundance and cover in determining habitat distribution in stream-dwelling cutthroat trout and invertebrate drift. Ecology, **67**: 898–911.

Winer, B.J. 1971. Statistical principles in experimental design. McGraw-Hill, New York.

Wiseman, S.W., Cooper, S.D., and Dudley, T.L. 1993. The effects of trout on epibenthic odonate naiads in stream pools. Freshwater Biol. **30**: 133–145.

Wooton, R.J. 1985. Energetics of reproduction. *In* Fish energetics: new perspectives. *Edited by* P. Tytler and P. Calon. Johns Hopkins University Press, Baltimore, Md. pp. 231–254.

© 1997 NRC Canada

Bayesian stock assessment using catch-age data and the sampling – importance resampling algorithm

Murdoch K. McAllister and James N. Ianelli

Abstract: A Bayesian approach to fisheries stock assessment is desirable because it yields a probability density function (pdf) of population model parameters. This pdf can help to provide advice to fishery managers about the consequences of alternative harvest policies and convey uncertainty about quantities of interest such as population biomass. In stock assessment, catch-age data are commonly used to estimate population parameters. However, there are few catch-age analyses that use Bayesian methods. In this paper, we extend the sampling – importance resampling algorithm so that a pdf of population model parameters can be estimated using catch-age data and indices of relative abundance. We illustrate the procedure by estimating a 54-parameter pdf for yellowfin sole (*Limanda aspera*) in the eastern Bering Sea. The example demonstrates how catch-age data can markedly improve Bayesian estimation, and also illustrates the potential for significant biases in treating trawl survey abundance indices as absolute measures of stock size.

Résumé : Une approche bayésienne à l'évaluation des stocks de poissons exploités par la pêche est souhaitable parce qu'elle donne une densité de probabilité (dp) des paramètres du modèle de population. Cette dp peut aider les gestionnaires des pêches à cerner les conséquences d'autres politiques de récolte et véhiculer l'incertitude au sujet des quantités d'intérêt, comme la biomasse de la population. Toutefois, il existe peu d'analyses des prises par âge qui font appel aux méthodes de Bayes. Dans le présent article, nous étendons l'algorithme échantillonnage – rééchantillonnage d'importance de sorte que l'on puisse estimer une dp des paramètres du modèle de population en utilisant les données sur les prises par âge et les indices d'abondance relative. Nous illustrons la méthode en estimant une dp à 54 paramètres pour la limande à nageoires jaunes (*Limanda aspera*) dans l'est de la mer de Bering. L'exemple démontre comment les données sur les prises par âge peuvent servir à améliorer grandement l'estimation bayésienne et, également, illustre la possibilité d'introduire des biais significatifs lorsqu'on traite les indices d'abondance obtenus à l'aide de relevés par chalutage comme étant des mesures absolues de la taille des stocks. [Traduit par la Rédaction]

Introduction

Many of the models used to provide advice to fishery managers, for example, about total allowable catches (TACs), require fixed values for input parameters (e.g., natural mortality rate (M), recruitment, and fishery selectivity). These assumptions often mask the level of uncertainty inherent in stock assessments. Bayesian statistical approaches have recently been applied to incorporate such uncertainties in advice to fishery managers (Punt 1993; McAllister et al. 1994; Stocker et al. 1994; Walters and Ludwig 1994; Walters and Punt 1994; Ianelli and Heifetz 1995; Raftery et al. 1995; Kinas 1996). The resulting Bayesian probability distributions can readily convey to fishery managers the uncertainty in derived model quantities such as the biomass of a fish population. Bayesian methods can also help managers to assess the likelihood of a variety of outcomes, e.g., rates of population decline, that could result from a proposed management action such as setting a harvest level or TAC.

A drawback of adapting Bayesian methods to large (many parameter) nonlinear models such as those used for age-structured fisheries stock assessment has been with calculating the multidimensional integrals that are fundamental to the joint probability distribution of model parameters. Monte Carlo methods are the most common methods for approximating such high-dimensional integrals. These include Markov Chain Monte Carlo methods (e.g., Metropolis–Hastings algorithm) and importance sampling (e.g., sampling – importance resampling (SIR) algorithm) (Metropolis et al. 1953; Hastings 1970; Berger 1985; Rubin 1987, 1988; Gelfand and Smith 1990; Smith 1991; West 1993). We first provide a brief overview of the application of these methods in stock assessment. We then suggest an extension of one of these methods so that catch-age data can be used in addition to relative abundance data.

Of the Monte Carlo methods, the SIR algorithm (Rubin 1987, 1988) has been applied most often in Bayesian fisheries assessments (e.g., Punt 1993; McAllister et al. 1994; Stocker et al. 1994; Raftery et al. 1995). In a study that was not explicitly Bayesian, Francis et al. (1992) used a method analogous to the SIR algorithm to calculate the biological

Received January 23, 1996. Accepted July 31, 1996.
J13266

M.K. McAllister.[1] Fisheries Research Institute, Box 357890, University of Washington, Seattle, WA 98195-7980, U.S.A.
J.N. Ianelli. National Marine Fisheries Service, Alaska Fisheries Science Center, 7600 Sand Point Way NE, Seattle, WA 98115, U.S.A.

[1] Present address: Renewable Resources Assessment Group, Imperial College, 8 Princes Gardens, London, SW7 1NA, U.K.

© 1997 NRC Canada

risks of alternative harvesting policies for orange roughy (*Hoplostethus atlanticus*). Only two of several types of the population model input parameters used (i.e., average unfished biomass, B_0, and recruitment anomalies, ϵ_y) were treated as uncertain. Raftery et al. (1995) used the SIR algorithm to estimate probability distributions for bowhead whale (*Balaena mysticetus*) population parameters. The procedure used considerable prior information on input parameters for a deterministic population dynamics model and estimated quantities relating to the current status of the population such as the replacement yield in the current year. McAllister et al. (1994) generalized these approaches to provide the flexibility to incorporate uncertainty in recruitment deviates about a stock–recruit function, informative priors for several model parameters, and a Bayesian decision analysis of alternative harvesting policies. They estimated the posterior by fitting an age-structured population dynamics model to relative indices of abundance and using age-structured data to construct priors on recruitment anamolies, ϵ_y. However, there is often relatively little information about input parameters to population dynamics models in relative abundance indices. In contrast, the incorporation of age frequency or catch-age data from the commercial catch and (or) from research surveys to the likelihood function can help to improve statistical inference about input parameters and derived quantities such as stock biomass.

In this paper, we extend the use of the SIR algorithm so that a population dynamics model can be fitted to fishery catch-age data and survey data. We illustrate the algorithm using eastern Bering Sea yellowfin sole (*Limanda aspera*) data. In the example, we present a new procedure to construct a prior probability distribution for the constant of proportionality for trawl survey abundance indices that uses survey data and expert judgment. The example illustrates the potential improvements in statistical inference that can be obtained by including catch-age data in a Bayesian stock assessment. The example also illustrates the large biases that can result from using area-swept survey abundance indices as absolute rather than relative measures of stock size.

A conceptual framework for a Bayesian stock assessment

McAllister et al. (1994) defined a conceptual framework for a Bayesian stock assessment: a population dynamics model is fitted to data to construct a posterior probability density function (pdf) of the vector of model parameters, θ, (e.g., B_0, M, ϵ_y). A "state of nature" is one potential realization of the set of all possible values for θ, say θ_i. The probability that a given state of nature θ_i is true conditioned on the data, $P(\theta_i \mid \text{data})$, is given by

$$(1) \qquad P(\theta_i \mid \text{data}) = \frac{L(\text{data} \mid \theta_i)p(\theta_i)d\theta}{\int L(\text{data} \mid \theta)p(\theta)d\theta},$$

where $p(\theta_i)$ is the prior probability density for state of nature i and $L(\text{data} \mid \theta_i)$ is the likelihood function for the data evaluated at θ_i. The value $p(\theta)$ represents the probability that a given state of nature is true prior to obtaining a set of data that can further our ability to discriminate among alternative θ_i.

Using sampling – importance resampling and catch-age data to construct probability distributions for quantities of interest

A sample from the posterior distribution, $P(\theta \mid \text{data})$, can be used to estimate marginal probability distributions for quantities of interest, such as current population biomass, that are known functions of the parameters, θ. This sample can be drawn using the SIR algorithm where it is possible to generate a sequence of i.i.d. (independent and identically distributed) random variables $(\theta_1, \theta_2, \ldots, \theta_m)$ that have a common probability density function, $h(\theta)$, called an importance function (Berger 1985). McAllister et al. (1994) summarized the statistical basis for the SIR algorithm. The procedure requires that for each vector θ_k that is drawn from $h(\theta)$, an importance ratio is calculated:

$$(2) \qquad w(\theta_k) = \frac{L(\text{data} \mid \theta_k)p(\theta_k)}{h(\theta_k)}.$$

The assignment of importance ratios forms a discrete distribution $F(\theta \mid \text{data})$ over $(\theta_1, \theta_2, \ldots, \theta_m)$ placing mass

$$(3) \qquad F(\theta_k \mid \text{data}) = \frac{w(\theta_k)}{\sum_{k=1}^{m} w(\theta_k)}$$

on each θ_k. The distribution, $F(\theta \mid \text{data})$ over $(\theta_1, \theta_2, \ldots, \theta_m)$, approximates the actual posterior distribution, $P(\theta \mid \text{data})$. This approximation improves as m increases (Berger 1985, p. 263; McAllister et al. 1994).

To estimate the marginal posterior distribution for a quantity of interest that is a known function of θ (e.g., $g(\theta)$), the discrete distribution $F(\theta \mid \text{data})$ over $(\theta_1, \theta_2, \ldots, \theta_m)$ could be used (i.e., each $g(\theta_k)$ would be assigned the mass $F(\theta_k \mid \text{data})$ over $(\theta_1, \theta_2, \ldots, \theta_m)$) (Berger 1985, p. 263). However, this may be numerically inefficient because m is often very large. Therefore, a random sample of size n, with $n < m$, is taken from $F(\theta \mid \text{data})$ over $(\theta_1, \theta_2, \ldots, \theta_m)$ (i.e., the probability of drawing $\theta_1, \theta_2, \ldots, \theta_m$ is proportional to $F(\theta_1 \mid \text{data})$, $F(\theta_2 \mid \text{data}), \ldots, F(\theta_m \mid \text{data})$). This step is the resampling part of the SIR algorithm. The empirical distribution for $g(\theta)$ is then given by $g(\theta_1), g(\theta_2), \ldots, g(\theta_n)$ with mass $1/n$ placed on each of the $g(\theta)$s.

A key task of importance sampling is that of finding a suitable $h(\theta)$ (van Dijk and Kloek 1983; Berger 1985). This task becomes increasingly challenging as the number of parameters increases, and the posterior becomes more complex (e.g., multimodal). Oh and Berger (1992) suggested the following three guidelines for selecting an importance function. (*i*) It should be easy to generate draws from. (*ii*) The tails of $h(\theta)$ (the value $h(\theta)$ at extreme values of θ) should not be less dense than the tails of the posterior; otherwise, estimated posterior expectations of quantities of interest may have large variance or even fail to converge with increasing m. (*iii*) The density function $h(\theta)$ should mimic the true posterior density function reasonably well (e.g., in skewness and covariance). If the data are not very informative, the model is fairly simple (e.g., deterministic), or adaptive importance sampling is used (see Discussion), the prior pdf can sometimes serve as an adequate importance function (Francis et

© 1997 NRC Canada

al. 1992; Kinas 1993; Punt 1993; McAllister et al. 1994; Raftery et al. 1995). However, in many instances, such as when catch-age data are available, the prior will be an inefficient importance function.

The multivariate Student t distribution, $t(\theta \mid d, \mu, V)$, has often been used as an importance function (van Dijk and Kloek 1983; Geweke 1989; West 1992; Kinas 1993):

$$(4)\qquad t(\theta \mid d, \mu, V) = \frac{\Gamma((d+p)/2)}{(\det V)^{1/2}(\pi d)^{p/2}\Gamma(d/2)} \times \left(1 + \frac{1}{d}(\theta - \mu)' V^{-1}(\theta - \mu)\right)^{-(d+p)/2},$$

where d is the degrees of freedom ($d > 0$), μ is the mean if $d > 1$, and V is a positive definite $p \times p$ matrix ($\mathrm{cov}(\theta) = dV/(d - 2)$ if $d > 2$) (from Berger 1985).

This distribution can be useful provided that each parameter in θ is defined on the interval $-\infty$ to ∞. An advantage of this distribution over the multivariate normal is that the tails of the distribution can be adjusted (by changing d) so that the density is slightly greater than the density in the tails of the posterior distribution. Furthermore, the mean, μ, can be estimated by the mode of the posterior by nonlinear minimization. V can be estimated by the negative inverse of the estimated Hessian matrix, i.e., the $p \times p$ matrix of second derivatives $(\partial^2 \log(P(\theta \mid \mathrm{data})))/(\partial\theta^i \partial\theta^j)$ where θ^i and θ^j are parameters i and j in θ, evaluated at the mode of $P(\theta \mid \mathrm{data})$ (van Dijk and Kloek 1983).

Steps for developing an importance function for use with age data

The following steps can be used to develop an importance function using the multivariate t density.

(1) Construct an informative prior for key model parameters such as natural mortality rate, M, and the coefficient of variation (CV) for recruitment variability, σ_r (see example below and Appendix 3).

(2) Construct likelihood functions for each of the types of data (see below).

(3) Use nonlinear minimization to find the posterior modal values for θ, $\theta_{P(\theta \mid \mathrm{data})}$. This is achieved by finding the values in the vector θ that minimize the quantity $-(\log(L(\mathrm{data} \mid \theta)) + \log(p(\theta)))$.

(4) Estimate the covariance, V, about the mode of the joint posterior by estimating the negative of the inverse of the Hessian matrix, $\hat{V}_{P(\theta \mid \mathrm{data})}$ (Arnold 1990).

(5) Set the importance function to $t(\theta \mid d, \hat{\theta}_{P(\theta \mid \mathrm{data})}, \hat{V}_{P(\theta \mid \mathrm{data})})$ where d is set to some value between 15 and 25 (values in this range have provided satisfactory results).

(6) Where appropriate, transform the parameters so that the importance function approximates the skewness in the posterior (van Dijk and Kloek 1983). If a transformation is used, the multivariate t density must be multiplied by the determinant of the Jacobian (van Dijk and Kloek 1983; Arnold 1990) to obtain the density function for the untransformed parameters, $h(\theta)$ (see example below and Appendix 1). This is necessary for calculating $w(\theta_k)$, since the terms of $w(\theta_k)$ are in θ, not in the transformed θ (eq. 2).

(7) Follow the standard steps for importance sampling (see below). This requires that parameter values be sampled or drawn from the importance function. We used the Choleski decomposition of the variance–covariance matrix to create parameter draws (Ripley 1987; Nash 1990; Kinas 1993).

(8) Modify the importance function if sampling from it appears to be too inefficient for estimating the posterior. A crude index of sampling efficiency is the maximum importance ratio, $w(\theta_k)$, taken as a proportion of the sum of all $w(\theta_k)$ over all of the draws. If the sample was taken from the posterior, this proportion would be equal to $1/m$, since each $w(\theta_k)$ would be equal to some constant. A target, for example, might be a maximum $w(\theta_k)$ of less than 5% of the total. However, this target may vary depending on the situation.

It is advisable to run replicate importance sampling trials with different random number sequences to evaluate whether the target can be consistently obtained and whether the replicate posteriors are sufficiently similar. In the example below, a maximum $w(\theta_k)$ of 4% of the total appeared to provide estimates of posterior pdfs sufficiently precise for stock assessment and decision analysis (see Results).

Another indicator of sampling efficiency is the CV in $w(\theta_k)$ where the CV is given by

$$(5)\qquad \mathrm{CV}(w(\theta_k)) = \frac{\mathrm{SD}(w(\theta_k))}{\frac{1}{m}\sum_{k=1}^{m} w(\theta_k)}$$

In the example below, increases in CVs to above 60 after about 50 000 draws of θ from $h(\theta)$ indicated that the importance function was inefficient and required adjustment. It is also useful to keep track of the parameter values in draws with the maximum $w(\theta_k)$ to compare with the modal estimates (Oh and Berger 1992). If the drawn values with maximum $w(\theta_k)$ are in the tails of the posterior distribution, the tails of the importance function may be too thin and need to be modified. This can be remedied by reducing the degrees of freedom, deemphasizing priors in estimating the Hessian matrix (see example below), or by reparameterizing to adjust for skewness. The split t distribution has been suggested as a more flexible distribution than the multivariate t for making adjustments for skewness (Geweke 1989). Importance sampling efficiency can also be improved by using estimates of the covariance matrix of the posterior distribution obtained from the first round of importance sampling in place of the Hessian estimate (van Dijk and Kloek 1983). This is because the estimate of posterior covariance provided by the Hessian matrix is a "local" estimate of covariance at the mode of the posterior; the alternative is a "global" estimate of the posterior covariance (van Dijk and Kloek 1983).

Steps of the SIR algorithm when age data are used

Below, we summarize the steps required to extend the SIR algorithm in McAllister et al. (1994) to the use of catch-age data from commercial catches and research surveys.

(1) Once an importance function, $h(\theta)$, has been chosen, draw one set of values θ_k for the model inputs from $h(\theta)$ (k refers to draw number k, since this step is repeated many times). For example, draw one set of values for B_0, α, M, q, s_{50}, a_{50}, and the recruitment residuals, ϵ_y, from $h(\theta)$ where α is a stock–recruit parameter, q is a catchability coefficient for

© 1997 NRC Canada

trawl survey estimates of biomass, s_{50} and a_{50} are selectivity parameters, and the other parameters have been defined above (see below).

(2) Project the population dynamics model from the initial year of the fishery forward to the current year (e.g., see below), using the values of model inputs, θ_k, drawn in step 2 and the known catches to predict stock biomass and catch-age data. Note that it is unnecessary to assume that unfished biomass is at equilibrium. Recruitment is assumed to be log-normally distributed about the assumed (e.g., Ricker) stock–recruit function (eq. 9). Values for all other parameters are assumed to be stationary over time. (The projection of the model from unfished conditions up to the current year will be referred to as historic trajectory k.)

(3) Evaluate the likelihood function, $L(\theta_k \mid \text{data})$, mass of the prior, $p(\theta_k)$, and mass of the importance function, $h(\theta_k)$, for the kth draw from $h(\theta)$, i.e., historic trajectory k (e.g., see below).

(4) If any of the observed catches exceeds the predicted exploitable biomass, set the prior to 0. This can be achieved by adding a penalty function to the log likelihood (see eqs. 20 and 21).

(5) Calculate the importance ratio, $w(\theta_k)$, for historic trajectory k (eq. 2).

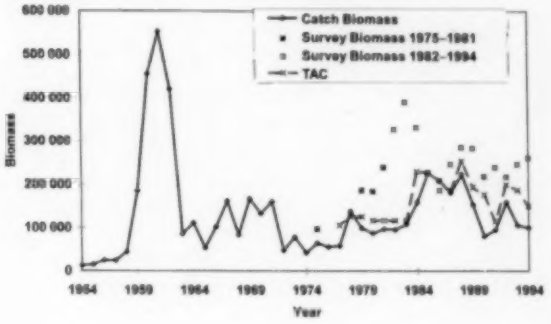
(6) Repeat steps 2–6 until the desired level of sampling precision is obtained. In the example below, $m = 3\,000\,000$ reduced the maximum $w(\theta_k)$ to 4% of the total. With a fast desktop computer (e.g., a Pentium 100) and programming language (e.g., C), this could potentially be achieved in an overnight run.

(7) Draw a large number (e.g., $n = 10\,000$) of θ_k randomly with replacement from $(\theta_1, \theta_2, \ldots, \theta_m)$ with probabilities proportional to $w(\theta_1), w(\theta_2), \ldots, w(\theta_m)$. The resulting sample $(\theta_1, \theta_2, \ldots, \theta_n)$ approximates an i.i.d. sample from the joint posterior pdf of θ. This sample could be used when performing the decision analysis (McAllister and Pikitch 1997). It is also useful for estimating the marginal posterior probabilities and covariances for model input parameters (McAllister et al. 1994).

Estimating a joint posterior probability distribution of population dynamics model input parameters for yellowfin sole in the eastern Bering Sea

The exploitation history of yellowfin sole in the eastern Bering Sea extends back to the mid 1950s. In the early 1960s, the population was heavily exploited by foreign trawl vessels with annual catches up to about 550 000 t (Wilderbuer et al. 1992) (Fig. 1). Since the early 1970s, fishery removals have dropped off to about 100 000 t annually and recruited biomass appears to have recovered to levels over 2 million t (Wilderbuer et al. 1992). The current U.S. domestic fishery is managed by TAC quotas that are set annually. Assessment scientists provide the estimates of stock biomass and allowable biological catch (ABC). The ABC is obtained by applying to the catch equation the estimate of fishing mortality rate that will reduce spawner biomass per recruit to 35% of that under unexploited conditions, $F_{35\%}$ (Clark 1991). The assessment typically uses age-structured data from the commercial fishery and a trawl survey and, also, absolute

Fig. 1. Catch biomass (t), trawl survey (t/10), and TAC (t) series for yellowfin sole in the eastern Bering Sea.



estimates of biomass from the trawl survey. However, controversy exists over abundance estimates because recent gear efficiency studies (D. Somerton, Alaska Fisheries Science Center (AFSC), National Marine Fisheries Service (NMFS), Seattle, Wash.) indicate that, for some species, trawl survey gear is considerably more efficient than is commonly assumed. Below, we describe the model that we used to assess the status of yellowfin sole and the specifications used to estimate a joint posterior probability distribution for the model parameters and other quantities of interest for fisheries management.

Population dynamics model

The model is age-structured and assumes that annual recruitment is lognormally distributed about a Ricker stock–recruit function. The model assumes that recruitment to the fishery follows a logistic function of age such that a fixed fraction of individuals within an age class will recruit annually to the fishery from individuals not previously vulnerable to fishing. In each year, fishing mortality is assumed to occur after natural morality has occurred. In 1954, the initial year of the fishery, population biomass is assumed to be distributed about B_0 and age structure is partially stationary ($\epsilon_y = 0$ for ages >8 years).

Resource dynamics

The dynamics of animals of age 2 and above are governed by the equation

$$
\begin{aligned}
N_{y+1,a} &= e^{-M}(N_{y,a-1}(1-H_y) + U_{y,a-1}\tau_a) && 2 \le a < m \\
U_{y+1,a} &= e^{-M}U_{y,a-1}(1-\tau_a) && 2 \le a < m \\
N_{y+1,m} &= e^{-M}(N_{y,m} + N_{y,m-1})(1-H_y) && a = m
\end{aligned} \tag{6}
$$

where $N_{y,a}$ is the number of recuited animals of age a at the start of the year y, $U_{y,a}$ is the number of unrecruited animals of age a at the start of year y, τ_a is the proportion of unrecruited animals of age $a-1$ that recruit at age a:

$$\tau_a = (\phi_a^f - \phi_{a-1}^f)/(1-\phi_{a-1}^f), \tag{7}$$

ϕ_a^f is the fraction of animals of age a that would be recruited

© 1997 NRC Canada

if the population were at its deterministic unexploited equilibrium level:

$$\phi_a^f = \frac{1}{1+\exp(-s_{50}^f(a-a_{50}^f))}, \tag{8}$$

s_{50}^f is the slope of the fishery recruitment ogive at 50% recruited, a_{50}^f is the age at 50% recruited, M is the instantaneous rate of natural mortality on animals, H_y is the harvest rate during year y and occurs after natural mortality (all recruited animals are assumed to be equally vulnerable to fishing), and m is the maximum (lumped) age-class (all animals in this and the previous age-class are recruited and mature).

Births

$$\begin{aligned} N_{y,1} &= 0 \\ U_{y,1} &= (E_{y-1}\exp(\alpha_0(1-E_{y-1}/\beta)))e^{\epsilon_y - I_y\sigma_r^2/2}, \end{aligned} \tag{9}$$

where E_y is the total reproductive potential of the population during year y at the time of spawning:

$$E_y = e^{-M}\sum_{a=1}^{m}\mu_a f_a(N_{y,a}(1-0.5H_y)+U_{y,a}), \tag{10}$$

μ_a is the fraction of mature fish at age a (see Table 3), f_a is the reproductive potential per individual fish of age a, and is assumed equal to w_a, w_a is the mass of a fish of age a (assumed to be constant throughout the year) (see Table 3):

$$w_a = \delta_1(L_a)^{\delta_2}, \tag{11}$$

$L_a = L_\infty(1-e^{-k(a-t_0)})$, ϵ_y is the recruitment residual for year y: $e^{\epsilon_y - I_y\sigma_r^2/2}$ is referred to as the recruitment multiplier ($\epsilon_y \sim$ Normal(0, σ_r^2) (see Table 2), I_y is set to 1 if the model is used for decision analysis (McAllister and Pikitch 1997) and the year is greater than the last year in which ϵ_y is estimated (i.e., 1990) and 0 otherwise (setting I_y to 1 activates the lognormal correction error factor; thereby the expected value for recruits, $E[U_{y,1}]$, equates with the value given by the stock–recruit function), σ_r is the standard deviation of the log of the multiplicative fluctuations in births, and α_0, β are stock–recruit function parameters.

Initial conditions

The initial numbers-at-age (in 1954 for yellowfin sole) are given by the equation

$$\begin{aligned} N_{y_1,a} &= R_1\phi_a\exp(-(a-1)M)e^{\epsilon_a} && 1 \le a \le m-1 \\ N_{y_1,m} &= R_1\exp(-(m-1)M)/ \\ &\quad (1-\exp(-M)) && a = m \\ U_{y_1,a} &= R_1(1-\phi_a) \\ &\quad \times\exp(-(a-1)M)e^{\epsilon_a} && 1 \le a \le m-1 \end{aligned} \tag{12}$$

where R_1 is the number of 1 year olds at the deterministic equilibrium that corresponds to an absence of harvesting and ϵ_a is estimated only for cohorts that are included in the first year of age data. For yellowfin sole, catch age data start in 1964 and the plus group for the age data is 18 yr. Therefore, the maximum age in which ϵ_a is estimated was age 8. For $a > 8$, ϵ_a was set to 0. A value for R_1 is calculated from the value for the virgin biomass at midyear (after natural mortality), B_0, using the equation

$$R_1 = B_0/\left\{\sum_{a=1}^{m-1} w_a\phi_a^f\exp(-aM) + w_m\exp(-mM)/[1-\exp(-M)]\right\}. \tag{13}$$

Values for the stock–recruit function parameters α and β are calculated from the values of R_1:

$$\begin{aligned} \alpha_0 &= \alpha + 7M \\ \beta &= \frac{\tilde{B}_0^S R_1}{(1-\log(1/\tilde{B}_0^S)/\alpha_0)} \\ \tilde{B}_0^S &= \left(\sum_{a=1}^{m-1}\mu_a f_a e^{-aM} + \mu_a f_a \times\exp(-mM)/(1-\exp(-M))\right), \end{aligned} \tag{14}$$

where $\tilde{B}_0^S$ is the reproductive potential per recruit and α is the value for α that is selected from the prior distribution for α. α is given in units of individual age 7 recruits after natural mortality. The adjustment of α to α_0 is made to rescale the prior value to the units for recruits in the assessment model (in age 1 recruits before natural mortality).

Catches

The exploitation rate during year y, H_y, is obtained by solving the equation

$$H_y = \frac{C_y}{B_y}, \tag{15}$$

where C_y is the observed annual catch biomass during year y (assumed to be observed without error) and B_y is the exploitable biomass in year y:

$$B_y = \sum_{a=1}^{m} w_a N_{y,a} e^{-M}. \tag{16}$$

Types of data used

The population dynamics model was fitted to two series of biomass estimates from the trawl surveys in the eastern Bering Sea: the first series is from 1975 to 1981 and the second from 1982 to 1994 (Table 1). The data were provided by G. Walters (AFSC, NMFS, Seattle, Wash.). The two series were treated separately because the trawl survey gear was changed in 1982 (see Gunderson 1993 for details). Both series were treated as indices of relative abundance, each with its own constant of proportionality. Both were assumed to have the same asymptotic selectivity function because the fit of the model to the data was not improved with separate selectivity estimates. The removals by survey gear were assumed to have an insignificant effect on stock size and to have occurred after recruitment and natural mortality and after half of the commercial catch had been removed.

© 1997 NRC Canada

Table 1. Total commercial catch biomass and trawl survey estimates of biomass of yellowfin sole in the eastern Bering Sea (tonnes).

Year	Catch	Survey	Year	Catch	Survey
1954	12 562	—	1975	64 690	972 500
1955	14 690	—	1976	56 221	—
1956	24 697	—	1977	58 373	—
1957	24 145	—	1978	138 433	—
1958	44 153	—	1979	99 017	1 866 500
1959	185 321	—	1980	87 391	1 842 400
1960	456 103	—	1981	97 301	2 394 700
1961	553 742	—	1982	95 712	3 275 300
1962	420 703	—	1983	108 385	3 910 600
1963	85 810	—	1984	159 526	3 320 300
1964	111 177	—	1985	227 107	2 277 400
1965	53 810	—	1986	208 597	1 866 400
1966	102 353	—	1987	181 428	2 465 800
1967	162 228	—	1988	223 156	2 854 600
1968	84 189	—	1989	153 170	2 832 180
1969	167 134	—	1990	80 584	2 183 800
1970	133 079	—	1991	95 000	2 393 300
1971	160 399	—	1992	159 038	2 172 900
1972	47 856	—	1993	106 101	2 465 400
1973	78 240	—	1994	100 000	2 610 481
1974	42 235	—	1995	100 000	—

Note: The CV in the survey biomass data was 0.19 (obtained using equations and data in McAllister 1995, Chap. 1).

The trawl survey data are assumed to be proportional to the biomass that is susceptible to the survey gear (i.e., B'_y):

$$(17)\qquad B'_y = e^{-M}\sum_{a=1}^{m}\phi'_a(N_{y,a}(1-0.5H_y)+U_{y,a})w_a,$$

where ϕ'_a is the proportion of individuals at age a that are vulnerable to the survey gear,

$$(18)\qquad \phi'_a = \frac{1}{1+\exp(-s'_{50}(a-a'_{50}))},$$

s'_{50} is the slope of the selectivity ogive at 50% selected, and a'_{50} is the age at 50% selected.

Both series of abundance indices are treated as being log-normally as

$$(19)\qquad \log(O^i_j) = \text{Normal}(\log(q_iE^i_j),\sigma^2_i),$$

where O^i_j is the jth value of the ith series, q_i is the constant of proportionality of the ith series, E^i_j is the model estimate of the quantity corresponding to O^i_j (e.g., B'_y in eq. 17), and α_i is the coefficient of variation for series i. The contribution of index i to the log likelihood is

$$(20)\qquad \lambda^i = -0.5n_i\log(2\pi) - \sum_j \log(\sigma_iO^i_j) - \frac{0.5}{\sigma^2_i}\sum_j \log(O^i_j/(q_iE^i_j))^2,$$

where n_i is the number of data points for abundance index series i. The first two terms were ommitted from the calculations because they contain constants only. The values for the parameters in $q_iE^i_j$ are treated as unknown and estimated.

The proportions at age in the commercial catch are assumed to be given by

$$(21)\qquad c^f_{a,y} = \frac{N_{a,y}}{\sum_{a'=1}^{m} N_{a',y}}$$

The proportions at age in the trawl survey are assumed to be given by

$$(22)\qquad c'_{a,y} = \frac{\phi'_a((1-0.5H_y)N_{a,y}+U_{a,y})}{\sum_{a'=1}^{m}\phi'_{a'}(1-0.5H_y)N_{a',y}+U_{a',y})}$$

In both the commercial catch-age data series (extending from 1964 to 1993) and the survey data series (1975 to 1993) (Table 1), the observed numbers at age in each year were assumed to be multinomially distributed. The contribution of series i to the log likelihood is

$$(23)\qquad \lambda^i = \hat{T}^i\sum_{j=1}^{n_i}\sum_{a=1}^{m'} o^i_{a,j}\log(c^i_{a,j})$$

where $\hat{T}^i$ is the estimated effective sample size for the total number of fish sampled in each year, $o^i_{a,j}$ is the proportion at age from the original sample, $c^i_{a,j}$ is the predicted proportion of fish at age a in the jth year of series i, n_i is the number of years in series i, and m' is the plus group for the age data (18 yr for both series) (Appendix 2).

The number of fish that were effectively sampled at random in each age sample (i.e., effective sample size, Pennington and Vølstad 1994) was estimated to be 102 for the fishery series and 104 for the survey series (see Appendix 2). In the estimation, the values used in the multinomial likelihood function for each year were rounded to 100.

To fit an age-structured model to catch biomass data series, we assumed that the fraction of fish harvested in each recruited age class is equal to the ratio of the observed catch biomass to the predicted exploitable biomass and that the catch biomass series is known without error (Francis 1992; Francis et al. 1992; Punt et al. 1993; McAllister et al. 1994).

The log likelihood also incorporated a term for H_y that sets the prior equal to 0 when the harvest rate exceeds the maximum possible (similar to the likelihood function of the observed catch biomass values in Bence et al. 1993):

$$(24)\qquad \lambda^5 = -0.5n_5\log(2\pi) - \sum_j \log(\sigma_5C_j) - \frac{0.5}{\sigma^2_5}\sum_j \log(C_j/\hat{C}_j)^2,$$

where σ_5 is the standard deviation in C_j (set very small, e.g., to 0.0002, and C_j are assumed to be measured without error), and $\hat{C}_j$ is given by:

$$(25)\qquad \hat{C}_j = C_j \text{ if } H_y < 0.99$$
$$\hat{C}_j = 0.99B'_y \text{ if } H_y \geq 0.99.$$

© 1997 NRC Canada

Prior probabilities used

The procedure used a joint prior pdf of multiple model input parameters. For simplicity, the prior assumes that the parameters are all independent of each other. For example, prior to evaluating the data, no combinations of values for B_0 and M are assumed to be more or less credible than implied by the product of their respective prior pdfs. Thus, under the assumption of independence, prior pdfs are constructed individually for each parameter and the joint prior is the product of the priors for each parameter:

$$(26) \qquad p(\theta_k) = \prod_{i=1}^{54} p(\theta_k^i),$$

where $p(\theta_k^i)$ is the prior for parameter i in the vector θ_k.

The 54 estimated model input parameters included the following: (*i*) virgin biomass, B_0, (*ii*) stock productivity, α (reflecting the slope parameter of the Ricker stock–recruit function at 0 spawners, eq. 9), (*iii*) natural mortality rate, M, (*iv*) the constant of proportionality, which relates the trawl estimates of stock biomass to actual biomass, q_i, (*v*) the age at 50% recruited, a_{50}^f, (*vi*) the slope of the fishery recruitment ogive at 50% recruited, s_{50}^f, (*vii*) the age at 50% vulnerable to the trawl survey, a_{50}^t, (*viii*) the slope of the trawl selectivity ogive at 50% vulnerability to trawl survey gear, s_{50}^t, and (*ix*) the annual lognormal recuitment residuals ϵ_y from the years 1946 to 1990 (eq. 9). The values for ϵ_y for the years 1934–1945 were fixed at 0 to provide reasonably efficient estimates of the posterior (the first year of reported catches is 1954). This caused an underestimate of the posterior variance for quantities such as B_0. However, the Hessian estimates of the CV in the marginal posterior distributions were only slightly less than when ϵ_y for the years 1934–1990 were estimated (e.g., by 1.6% for B_0).

The procedure uses informative prior pdfs of parameters B_0, α, σ_r, M, and q_i (Table 2). These priors were constructed from stock–recruit data for other flatfishes and by consulting the literature and soliciting expert judgment. The prior for B_0 is uniformly distributed over the interval 500–5000 kt. The priors for α and σ_r were constructed using an approach in McAllister et al. (1994). The prior for α is Normal(1.52, 0.78^2) and was obtained from stock and recruit data for 14 other populations of small-sized flatfishes (Fig. 2; and see Appendix J in McAllister 1995). The prior for σ_r is lognormal with a mean of 0.46 and an SD of 0.40 (Fig. 2; and see Appendix J in McAllister 1995). σ_r was fixed at the posterior modal estimate (0.40, obtained using nonlinear minimization) for estimating the posterior distribution with SIR because SIR was too inefficient when the prior for σ_r was used. McAllister et al. (1994) found that the estimate of the posterior was relatively insensitive to using an informative prior for σ_r versus the best estimate for the population. The prior pdf of M (Table 2) is lognormal and was parameterized by setting the 2.5 and 97.5 percentiles of the distribution to 0.08 and 0.18, a range consistent with knowledge of the life history of yellowfin sole, aging studies, and values used in stock assessments of 15 other populations of small-sized flatfishes (i.e., 0.10 to 0.12) (Wilderbuer et al. 1992; Myers et al. 1993).

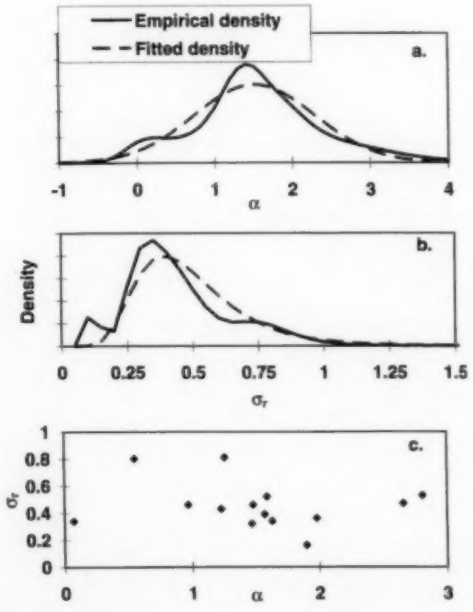
The prior pdfs of q_i were lognormal with a mean of 0.44 and a CV of 0.59 for the first series and a mean of 0.97 and a CV of 0.51 for the second series (Table 2). These prior pdfs were constructed by Monte Carlo simulation of the uncertainties in the following quantities: (*i*) the mean proportion of the population within the design in each year, (*ii*) the effective width swept by the net, as a fraction of the estimated width (this reflects the fraction of fish that encounter the net that are herded in from the trawl doors), (*iii*) the fraction that encounter the net that do not escape underneath the net, and (*iv*) the mean proportion of the fish detected as result of migration within the survey design and vessel avoidance (see Appendix 3 for details).

The priors for the selectivity parameters were diffuse but roughly centered about values consistent with previous as-

Table 2. Priors of some population dynamics model parameters.

Parameter	Prior
B_0 (×1000 t)	Uniform(500, 5000)
α	Normal(1.52, 0.78^2)
σ_r	lognormal(0.46, 0.40^2)
ϵ_y ($y \in$ 1946–1990)	Normal(0, σ_r^2)
M(year^{-1})	$M \sim$ lognormal(0.12, 0.2^2)
$q_{75\text{-}81}$	lognormal(0.44, 0.59^2)
$q_{82\text{-}94}$	lognormal(0.97, 0.51^2)
a_{50}^f, a_{50}^t(years)	Normal(7.5, 3^2), $a_{50}^f > 0$, $a_{50}^t > 0$
s_{50}^f, s_{50}^t	Normal(1.5, 2^2), $s_{50}^f > 0$, $s_{50}^t > 0$

Fig. 2. (*a*) Empirical and fitted prior pdf of α in the Ricker stock–recruit function based on stock–recruit data for 14 flatfish stocks in Myers et al. (1993) (see Appendix J in McAllister 1995). (*b*) Empirical and fitted prior pdf of σ_r. (*c*) Plot of the modal estimate of σ_r versus α for the 14 flatfish stocks.



© 1997 NRC Canada

sessments. The priors for the slope at 50% selected for the commercial and the survey ogives were both Normal(1.5, 2^2). The priors for age (in years) at 50% selected for the commercial and survey ogives were both Normal (7.5, 3^2). Both sets of priors were truncated at 0. It is generally improper to use the results of one assessment to specify priors in another. However, the priors chosen are very flat and do not affect the results (see below).

Importance function used for yellowfin sole

The importance function that was used to estimate the posterior for yellowfin sole was constructed using the following procedure. First, the parameters B_0, σ_r (when used), M, s^i_{50}, and q_i were log transformed so that when drawn from the multivariate t distribution, the untransformed values would be skewed as they are in the posterior (see Appendix 1 for details). Second, a nonlinear minimization procedure, based on a gradient search method that uses numerical derivatives (Press et al. 1992), was used to find the mode of the posterior. Third, the Hessian matrix was estimated using numerical derivatives of the log of the posterior (i.e., the likelihood times the prior) at its mode. However, only the priors for M and ϵ_y were included in estimating the Hessian matrix. By eliminating the other priors from the posterior, the estimated covariance is slightly larger than that for the posterior with all of the priors included. This procedure reduces the chance of using an importance function with sharper tails than the posterior and improves the efficiency of SIR. The importance function used was a multivariate t distribution with 25 degrees of freedom, the mean given by the estimate of the mode of the posterior (with all of the priors), and the variance given by the negative inverse of the Hessian matrix.

For the stock assessment, the resampling procedure selected 10 000 of the 3 000 000 draws from the importance function. The base case application of the stock assessment technique used the relative abundance indices in Table 1, catch-age data from the commercial fishery and the survey (McAllister 1995), the catch biomass series (Fig. 1; Table 1), the priors (Table 2), and the biological and technological parameters (Table 3).

Bayesian estimation of population dynamics model parameters and stock status

Bayesian estimates of uncertainty in model input parameters and the status of yellowfin sole in the eastern Bering Sea are conveyed by the marginal posterior pdfs of the following quantities: (*i*) the model input parameters (e.g., B_0, α, M, q_i, s^i_{50}, a^i_{50}, ϵ_y), (*ii*) current-year (1995) stock biomass (B_{1995}), and (*iii*) the amount of depletion from unexploited conditions (B_{1995}/B_0). Tests were conducted to assess the sensitivity of the posteriors to the use of age data, the use of a multivariate t distribution developed from properties at mode of the posterior as an estimate of the posterior distribution (this would lessen the lengthy Monte Carlo steps required by SIR (e.g., from 3 000 000 to 10 000)), and the amount of variability in recruitment, σ_r. We wanted to examine how well a multivariate t distribution approximated the posterior given by SIR. The multivariate t with 25 degrees of freedom approaches a multivariate normal.

In the first test, only the relative biomass data were used, and the age data excluded. The prior pdf was used as the importance function and the selectivity parameters were set at the modal estimates from the baseline assessment. In the second test, the posterior from SIR was compared with a multivariate t with 25 degrees of freedom (eq. 4), parameter μ given by the posterior modal estimates of parameters (with M, B_0, q_i, and s^i_{50} on a log scale), and parameter V given by the Hessian estimate of covariance at the mode of the posterior. Distributions for derived variables such as current stock size were computed numerically by taking 10 000 random draws of θ from the multivariate t distribution, placing mass 1 / 10 000 on each drawn θ_k, computing the derived variable, $g(\theta)$, for each θ_k, and then summing the mass in the appropriate interval over $g(\theta)$. In the third test, the value of σ_r was increased from 0.40 to the highest value plausible, 0.55 (i.e., the 99.7th percentile of the marginal posterior for σ_r, based on the Hessian estimate of the posterior variance in σ_r when σ_r is estimated jointly with the other parameters).

Table 3. Values of the model parameters that are assumed to be known without error in the Bayesian assessment of yellowfin sole.

Parameter	Value
L_∞ (cm)	35.8
κ (year^{-1})	0.147
t_0 (years)	0.47
δ_1 (g/cm^3)	0.009 721 7
δ_2	3.0564
Plus group in model: m (years)	21
Plus group in age data: m' (years)	18
μ_1–μ_8	0.000
μ_9	0.075
μ_{10}	0.135
μ_{11}	0.270
μ_{12}	0.375
μ_{13}	0.415
μ_{14}	0.460
μ_{15}	0.475
μ_{16}	0.486
μ_{17}	0.490
μ_{18}–μ_m	0.500

Results

Using catch-age data markedly updates the priors for most model input parameters (Figs. 3–7; Table 4). The marginal posteriors for most of the population dynamics model parameters have considerably lower variance than the priors (Table 4). For the estimated recruitment residuals in the last few years of the historic time series, the posterior reflects the priors because there are fewer data in the last years (Fig. 6). The posteriors for current stock size and depletion are also highly peaked (Fig. 7). The 95% Bayesian confidence intervals (CIs) about the recruited and survey stock biomass are very tight, especially for the earlier years when very large catches were taken and the population was heavily depleted (Figs. 1 and 8). The CIs become broader in the late 1970s, despite the addition of survey biomass and age data, mainly because the catches of yellowfin sole decreased

© 1997 NRC Canada

Fig. 3. Marginal posterior pdfs and priors of (*a*) α, (*b*) B_0, and (*c*) *M*. The marginal pdfs are shown for the baseline case, and the case in which the posterior was approximated by a multivariate *t* distribution with 25 degrees of freedom (see text for details).

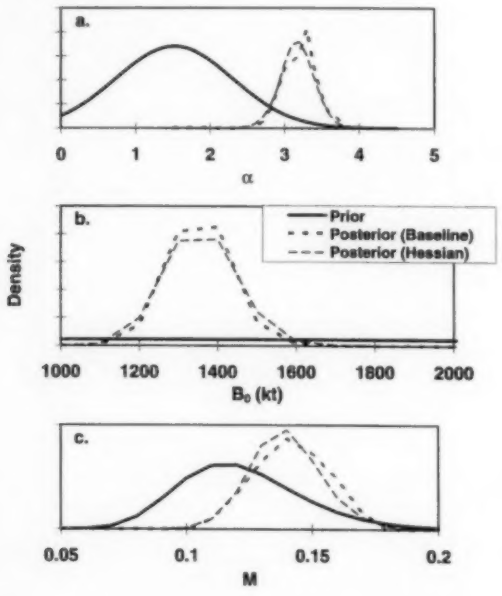


Fig. 4. Marginal posterior pdfs and priors of the catchability coefficient for the trawl survey for the periods (*a*) 1975–1981 and (*b*) 1982–1994. The marginals are shown for the baseline case and the case in which the posterior was approximated by a multivariate *t* distribution with 25 degrees of freedom (see text for details).

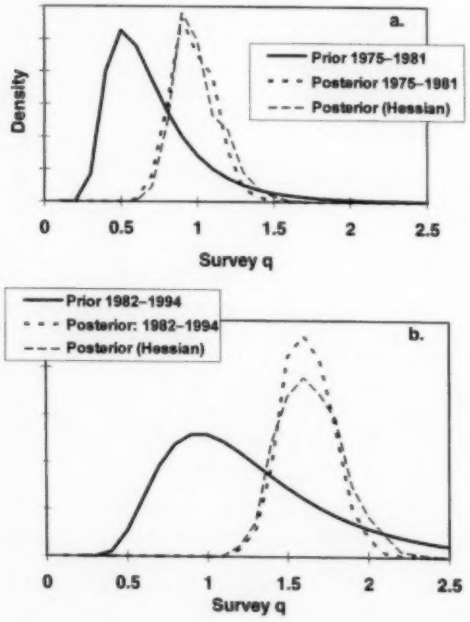


Fig. 5. Marginal posterior pdfs and priors of the (*a*) age at 50% selected and (*b*) slope of the logistic selectivity function at 50% selected for the fishery and the commercial catch. The marginals are shown for the baseline case only.

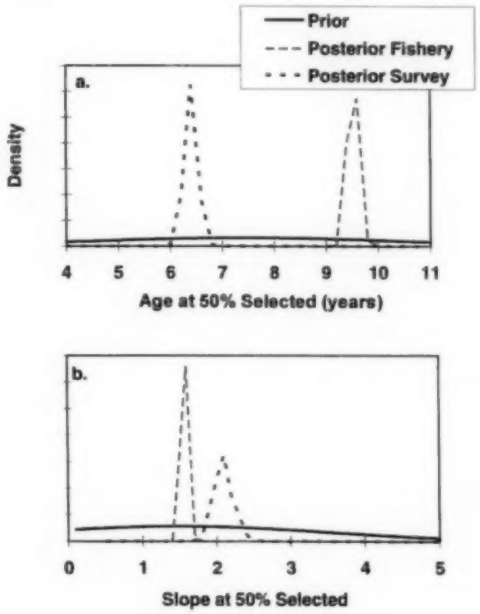
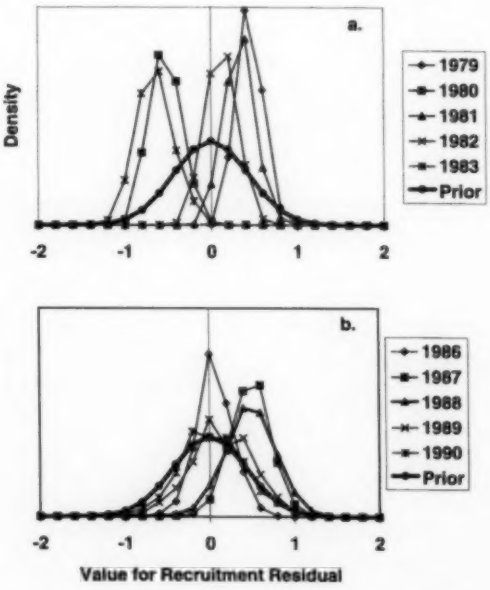


Fig. 6. Marginal posterior pdfs and priors of the recruitment residuals ϵ_y for the years (*a*) 1979–1983 and (*b*) 1986–1990. The marginals are shown for the baseline case only.



© 1997 NRC Canada

Table 4. Estimated ratio of the posterior variance to the prior variance for selected model input parameters.

Parameter	Baseline	Biomass data only	Age data only	$\sigma_r = 0.55$
B_0 (×1000 t)	0.005	0.507	0.006	0.006
α	0.082	0.550	0.078	0.111
M (year^{-1})	0.313	0.948	0.278	0.490
$q_{75\text{-}81}$	0.186	0.310	na	0.259
$q_{82\text{-}94}$	0.075	0.230	na	0.121
a^f_{50} (years)	0.001	na	0.001	0.001
a^s_{50} (years)	0.002	na	0.002	0.002
s^f_{50}	0.001	na	0.007	0.001
s^s_{50}	0.005	na	0.007	0.005
ε_{1985}	0.313	1.12	0.379	0.372

Fig. 7. Marginal posterior pdfs and priors of (*a*) virgin stock size (B_0), (*b*) biomass in 1995 (B_{1995}), and (*c*) stock depletion (B_{1995}/B_0). The marginals are shown for the baseline case with 3 million draws from the importance function and the three sequences of 1 million draws used and the case in which the posterior was approximated by a multivariaimates t distribution with 25 degrees of freedom (see text for details).

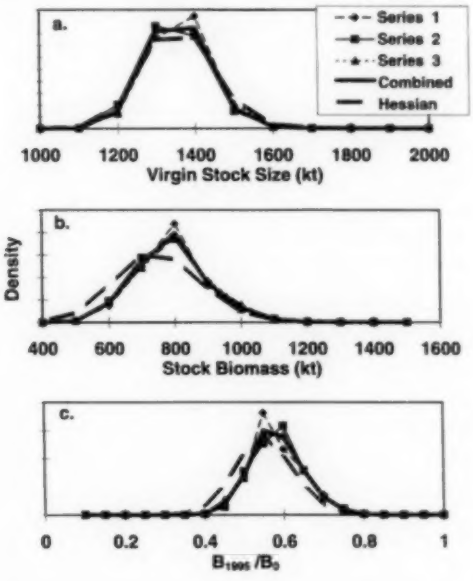
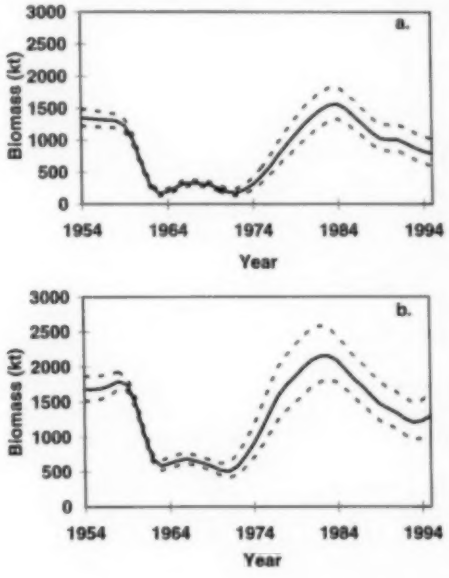


Fig. 8. Median and 95% confidence intervals for (*a*) recruited biomass and (*b*) surveyable biomass of yellowfin sole.



and inferences become less precise when fishing mortality rates are low (Bence et al. 1993). The plot of the modal estimates of recruitment versus spawning potential suggests a fairly well pronounced Ricker stock–recruit relationship with some of the largest recruitments occurring under the most depleted conditions (Fig. 9). However, there is still considerable scatter about the modal stock–recruit function as reflected by the modal recruitment CV (σ_r) of 0.40.

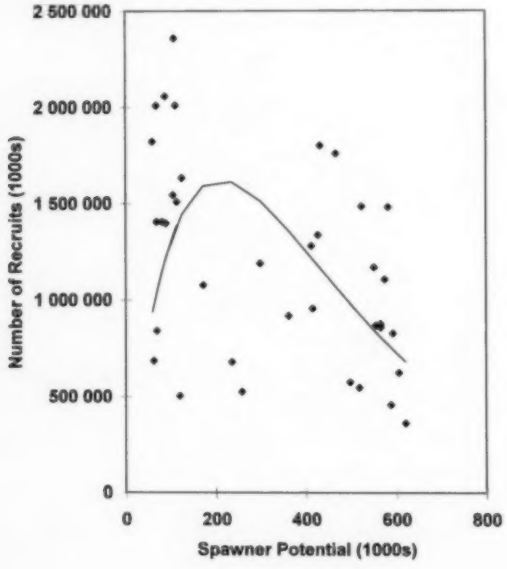
When only survey biomass data are used, the marginal posteriors for model input parameters are not much sharper than the priors and the marginal pdfs for quantities of interest such as current stock size are relatively flat (Figs. 10 and 11; Table 4). The survey biomass data suggest that stock size is higher and the stock less productive than under the baseline scenario when both age and biomass data are used.

The sampling efficiency in estimating the posterior was much greater when the model was fit to biomass data only. For example, the maximum $w(\theta_k)$ for a single draw from the importance function was consistently less than 1% of the total at less than 500 000 draws. In contrast, the maximum $w(\theta_k)$ was 4% after 3 million draws from the importance function when age data were also included (the maximum $w(\theta_k)$ were 12, 4, and 3% in each consecutive sequence of 1 million in the 3 million draws) (Table 5). See Discussion for comments on sampling efficiency.

In test 2, we compared posteriors obtained by using the SIR algorithm (called the SIR estimator) with a multivariate t based on the posterior modal parameter values for the mean, the Hessian estimate of the covariance of the posterior at the

© 1997 NRC Canada

Fig. 9. Modal estimates of the Ricker stock–recruit function and recruitment and spawner potential for the series 1954–1990.



mode, and degrees of freedom set at 25 (called the Hessian estimator, Figs. 3, 4, and 7). For most model input parameters, the Hessian estimator was reasonably close to the SIR estimator. However, the Hessian estimator favored smaller values for natural mortality rate (Fig. 3). For the derived quantities such as current stock size, the Hessian estimator was noticeably more pessimistic (Fig. 7). Current stock size is lower and depletion is more pronounced under the Hessian estimator. The differences in results between the two estimators appear to be due to real differences in the distributions rather than due to sampling variability. For example, the three replicate estimates of the posterior (of 1 million samples each) from SIR were much closer to each other than to the one estimated by the multivariate t (Fig. 7). Differences in marginals between the SIR estimator and the multivariate t do not necessarily imply that the posterior modal value for θ is different from that obtained by maximizing the posterior density function. The differences in estimated marginals between the posterior and the multivariate t could result from differences in their shape (e.g., the posterior could be banana-shaped, while the multivariate t is oval). Therefore, we conclude that at least for the yellowfin sole data analyzed here, the multivariate t with the posterior mode and Hessian estimate of covariance provides significantly poorer estimates of the posterior pdf relative to the SIR estimator.

The effect of replacing the value for σ_r of 0.40 with 0.55 was negligible: only slight changes in the marginal posterior pdfs of parameters and other quantities were observed. The variances in posterior pdf for model input parameters increased, as might be expected by increasing the amount of recruitment variability, but only slightly (e.g., the SD in B_0 under $\sigma_r = 0.55$ was 87 kt and under the baseline was 79 kt, see Table 4).

Fig. 10. Marginal posterior pdfs and priors of (*a*) α, (*b*) B_0, and (*c*) M. The marginals are shown for the baseline case and the cases in which $\sigma_r = 0.55$, and only the survey biomass data are used.

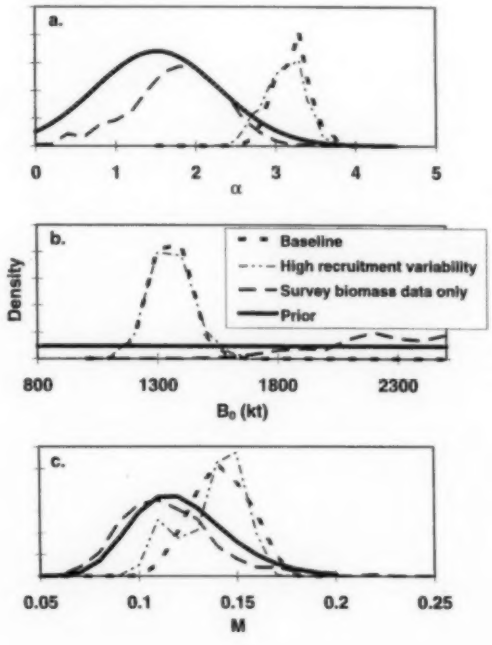
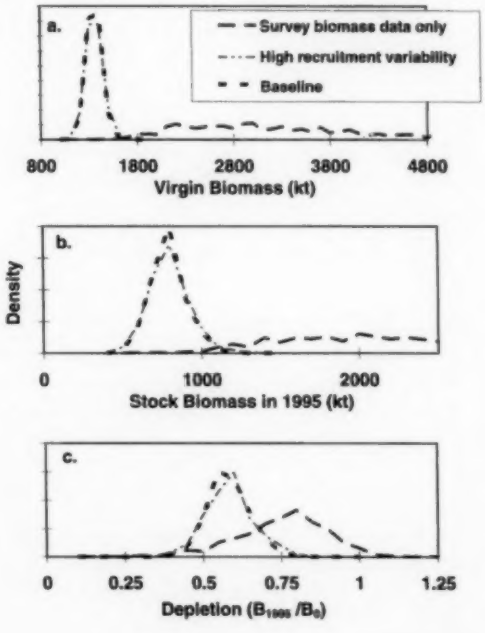


Fig. 11. Marginal posterior pdfs of (*a*) virgin stock size (B_0), (*b*) biomass in 1995 (B_{1995}), and (*c*) stock depletion (B_{1995}/B_0). The marginals are shown for the baseline case and the cases in which $\sigma_r = 0.55$, and only the survey biomass data are used.



© 1997 NRC Canada

Table 5. Posterior expectations and standard errors (in parentheses) for virgin biomass (B_0), current stock size (B_{1995}), and depletion (B_{1995}/B_0) for three replicate posteriors (each replicate was obtained by taking a series of 1 million i.i.d. draws from a multivariate t importance function; biomass values are in millions of tonnes).

Replicate	No. of draws	Max (% weight)	B_0	B_{1995}	B_{1995}/B_0
1	1 million	11.8	1.36 (0.08)	0.79 (0.10)	0.58 (0.06)
2	1 million	4.1	1.35 (0.08)	0.79 (0.11)	0.58 (0.07)
3	1 million	3.2	1.36 (0.08)	0.79 (0.11)	0.58 (0.07)
Combined	3 million	4.1	1.35 (0.09)	0.79 (0.11)	0.58 (0.07)

Discussion

This paper extends the use of the SIR algorithm and demonstrates that the use of catch-age data, in addition to relative abundance indices, for estimating a Bayesian posterior distribution can lead to markedly improved precision in estimates of population dynamics model parameters. The data series used in the example were highly informative because of the highly varied exploitation history of the population (i.e., high fishing effort followed by much lower effort, Hilborn and Walters 1992), the long time series of catch-age data, the existence of an independent survey of biomass and age frequency, and the high precision of the data series. For example, yellowfin sole is considered to be one of the most precisely aged fishes in the AFSC (Kimura and Lyons 1991).

However, the precision in posterior estimates is overestimated because several sources of uncertainty were ignored. These include uncertainty over the amount of recruitment variability (σ_r), the recruitment residuals, ϵ_y, for the earlier segment of the time series (1934–1945) (ages >8 years in 1954), mass at age, fecundity at age, catch biomass, the functional forms of survey and fishery selectivity and of the stock–recruit relationship, nonstationarity in model parameters, and autocorrelation in recruitment residuals. Furthermore, the multinomial likelihood function, often used in catch-age assessment methods (e.g., Methot 1990), may not adequately express the variability in the catch-age data. This is because sample units (e.g., trawl survey hauls) may capture groups of similarly aged individuals rather than individuals at random from the population, an assumption of the multinomial sampling process. However, in recent tests that we have conducted, estimated posterior variances were similar when the multivariate logistic likelihood function, derived specifically for catch-age data (Schnute and Richards 1995), replaced the multinomial. Even if additional uncertainties were effectively incorporated in a Bayesian assessment, the precision would still be considerably higher than that obtainable using abundance indices only.

In instances such as the current one, when data are highly informative, the issue of the choice of priors diminishes (Box and Tiao 1973). However, in the current procedure, informative priors are still needed for the amount of recruitment variability, σ_r, and the rate of natural mortality, M. These priors are necessary for the development of the importance function and to permit reasonably efficient SIR (McAllister et al. (1994).

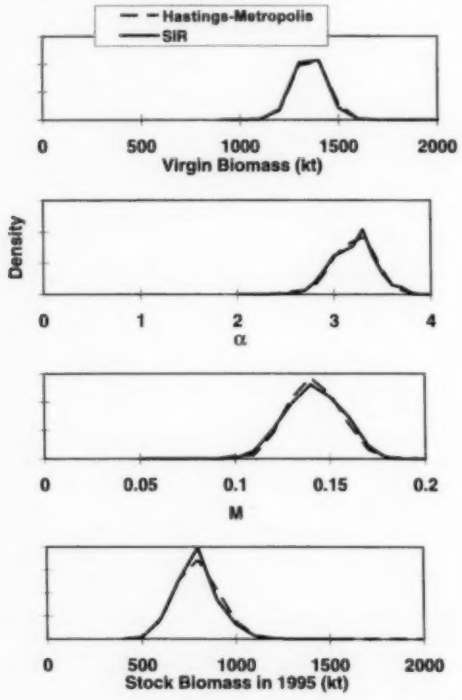
Alternative Monte Carlo methods for estimating posteriors in Bayesian fisheries assessments include adaptive importance sampling (AIS) and Markov Chain Monte Carlo (MCMC) methods (Givens 1993*a*, 1993*b*; Kinas 1993; A.E. Punt, Commonwealth Scientific and Industrial Research Organization (CSIRO), Hobart, Tasmania, Australia, personal communication). In contrast with SIR, AIS is an iterative importance sampling procedure that often uses as an importance function a finite mixture of multivariate pdfs, such as the multivariate Student density (Kinas 1993; West 1993). In AIS, the normalized importance ratios are used to develop a new importance function. This process is repeated until the importance function converges on the posterior distribution. A potential advantage of AIS over SIR is that AIS may be computationally more efficient than SIR, especially for estimating complex (e.g., multimodal) posteriors (Kinas 1993). However, AIS can be more difficult to implement because AIS is considerably more complicated than SIR. Furthermore, it is unclear whether AIS could deal effectively with the large number of uncertain parameters in catch-age analysis.

In contrast, MCMC methods are based on iterative Markovian updating schemes for estimating posteriors. "Sampling" entails a random walk over the posterior probability surface. MCMC methods can be easier to implement than SIR because they may require less coding and initial start-up time than importance sampling methods. Also, estimation of an importance function is not required. However, MCMC methods may be computationally less efficient than importance sampling methods (Smith 1991; Givens 1993*a*). For example, when we applied the Hastings–Metropolis version of the MCMC algorithm (Metropolis et al. 1953; Hastings 1970; Gelfand and Smith 1990) to the data for yellowfin sole, it took several times longer than the SIR algorithm to obtain a reasonably precise estimate of the posterior (Hastings–Metropolis procedure provided by A.E. Punt). Reassuringly, the distribution estimated by Hastings–Metropolis was very similar to that provided by SIR (Fig. 12). Tuning MCMC so that the draws taken are independent, however, is not straightforward (Raftery and Lewis 1992). In addition, there are some conditions in which the MCMC may not necessarily converge on the posterior (e.g., when the posterior surface is multimodal or not log-concave, Newton and Raftery 1994). Furthermore, testing for convergence is also not straightforward. Because of these various difficulties with implementation, it may be prudent to apply both the SIR and MCMC methods whenever time permits to test whether results agree.

As a further advantage, SIR output is more concise and versatile than MCMC output. SIR output for each sampled

© 1997 NRC Canada

Fig. 12. Marginal posterior and prior pdfs of (*a*) α, (*b*) virgin stock size (B_0), and (*c*) stock biomass in 1995. The marginals are shown for the baseline case estimated by SIR and the Hastings–Metropolis algorithms (see Discussion).



θ_k can be stored in two numbers: the initial random number seed that was used to generate each θ_k and the importance ratio, $w(\theta_k)$. These can easily be used to generate marginal posterior distributions of any quantity of interest in a stock assessment or decision analysis. Furthermore, it is unnecessary to rerun SIR to evaluate the sensitivity of functions of the posterior of θ (e.g., quantities from a decision analysis) to different priors. Instead, one simply divides each baseline $w(\theta_k)$ by the baseline prior $p(\theta_k)$ and multiplies by the new prior $p''(\theta_k)$ to give $w''(\theta_k)$. This avoids redoing the time-consuming steps of obtaining draws of θ_k from $h(\theta)$ and calculating the likelihood for each θ_k. The resampling step to obtain a set of draws from the new posterior of θ is then based on $w''(\theta_k)$ rather than $w(\theta_k)$.

In contrast, with MCMC, the full parameter vector must be stored for each sampled θ_k. Furthermore, to test the sensitivity of functions of the posterior of θ to different priors, it is necessary to rerun the MCMC procedure. This is because the random walk that produces the Markov Chain and the draws of θ from the posterior is a function of the likelihood and the prior. Therefore, the random walk must be reinitiated to obtain a set of draws of θ from a posterior that has a different prior. Despite these minor drawbacks, MCMC appears to be a promising alternative for Bayesian catch-age analysis. Further research on the application of MCMC (e.g., the Hastings–Metropolis algorithm) to catch-age data is required before an MCMC algorithm can be presented in detail as an alternative to SIR for Bayesian stock assessment and decision analysis.

When SIR is used, diagnostics are required to quickly evaluate whether the importance function chosen will provide a reasonably precise estimate of the posterior in an acceptable duration. They are also required to evaluate whether the estimated posterior is sufficiently close to the actual posterior. The diagnostics that we suggest are ad hoc but appear to work well in a variety of applications to catch-age data. In contrast, Kinas (1993) used the entropy relative to uniformity (ERU) which indicates how close the importance function is to the posterior. ERU is therefore not an absolute measure of sampling efficiency (i.e., it does not necessarily indicate whether an importance function will provide an acceptably precise estimate of the posterior or whether the final estimate of the posterior is sufficiently precise). For AIS, ERU is useful because a series of importance functions are used and the convergence of the importance function to the posterior can be observed. However, ERU is not appropriate for SIR because only one importance function is used. For example, there may be a large difference between the importance function and the posterior (i.e., ERU may be very low); but with many draws the final estimate of the posterior may be very precise.

We present the multivariate *t* distribution as an importance function that can be used for Bayesian catch-age analysis. However, in the example, this appears to be a relatively inefficient importance function because of the lengthy computing time, large number of draws used (3 million), and relatively high value for the maximum importance weight (i.e., 4%, 120 000 times higher than if the ideal importance function, the posterior, was used). This compares with 20 000 draws and a maximum weight <1% in McAllister et al. (1994) and about 300 000 draws and <1% in Raftery et al. (1995). Furthermore, it was not possible to include the prior for σ_r because SIR became too inefficient. Fixing σ_r at its modal value appeared to be a reasonable remedy and does not appear to bias the posterior (see McAllister et al. 1994). The SIR algorithm also performed poorly when all of the recruitment residuals at the beginning of the time series (the period during which no catch-age data exist) were treated as uncertain. Fixing the earliest residuals in the time series to zero remedied the situation and did not appear to bias the estimates of the posterior distribution.

Despite the inefficiencies of the multivariate *t*, an acceptably precise estimate of the posterior was obtained within a tolerable amount of computing time. For example, three replicate posteriors, each obtained by taking 1 million draws from the importance function, closely resemble each other (Fig. 7). Furthermore, the posterior expectations and standard errors for, e.g., B_0, B_{1995}, and B_{1995}/B_0 were very close despite the relatively large weights in each series (11.8, 4.1, and 3.2%) (Table 5). In other applications of the multivariate *t* distribution to catch-age data, much lower maximum weights have been obtained after fewer draws (e.g., <1% in <500 000 draws). Problems with efficiency could also be overcome by using better approximations of the posterior for the importance function or by using adaptive importance sampling (see Oh and Berger 1992; Kinas 1993; Newton and Raftery 1994).

Our approach differs in a variety of ways from that of Walters and Punt (1994), who used catch-age data to estimate a posterior distribution for the probability of sustaining current stock size from alternative allowable catch options.

© 1997 NRC Canada

For example, their estimation algorithm was based on virtual population analysis (backward calculation of numbers at age) tuned to a survey biomass series. Only few model parameters were treated as uncertain and a grid-based approach was used to compute the posterior distribution. Also, the posteriors on current-year stock biomass computed in Walters and Punt (1994) were more diffuse than the one in this paper because the time series of data was shorter and data were less informative.

Finally, the application of the procedure to yellowfin sole in the eastern Bering Sea demonstrates a simple way of incorporating uncertainty in scaling (via the q parameter) absolute survey abundance estimates. Clearly, uncertainty in this parameter can have broad implications for fishery management because the assessment of current stock size is the critical issue. For example, the estimates of population biomass given by the baseline Bayesian assessment were about 50% of those given with q fixed at 1.0 and assumed known with certainty. Also, by ignoring uncertainty in the value of q, the uncertainty in the estimate of current biomass will be underestimated. Hence, we advocate treating q as an estimated parameter and expressing stock assessment uncertainty using estimates of posterior probability distributions. Information about q (e.g., from studies on gear efficiency) that is not contained in the likelihood function can be included in the prior as has been done in the current paper. The idea that biases can result from treating q as known and fixing it at some value is not new but needs to be reemphasized because of existing practices.

The Bayesian estimation approach developed in the current paper is flexible and intuitively appealing and could be applied in a wide variety of situations involving catch-age data. For example, the procedure has been applied successfully to other sets of fishery age data in the Pacific Northwest (J. Ianelli), New Zealand (T. Hammond and R. Hilborn, School of Fisheries, University of Washington, Seattle, Wash.), England, and Australia (A.E. Punt, CSIRO, Hobart, Tasmania). The multivariate pdf that is estimated by the procedure is not only useful in stock assessment. It is also a key input to decision analyses of alternative management policies (Francis et al. 1992; McAllister et al. 1994). The posterior pdf of population parameters for yellowfin sole that we estimated is used in a companion paper (McAllister and Pikitch 1997) to evaluate the potential trade-offs among alternative trawl survey designs in the eastern Bering Sea.

Acknowledgments

This work was funded by the U.S. National Marine Fisheries Service. We thank Tom Wilderbuer and Gary Walters for providing survey and fishery data for yellowfin sole. We are grateful to Nick Barrowman and Ransom Myers for providing the stock and recruitment data for other flatfishes. We thank Ellen Pikitch, Judy Zeh, Ray Hilborn, Don Gunderson, Gary Stauffer, Geof Givens, and Tim Hammond for helpful discussions and comments on earlier versions of the manuscript. We thank Andre Punt for providing code for the Hastings–Metropolis algorithm, helpful discussions, and comments on the manuscript. We thank David Somerton and Craig Rose for their assistance in developing informative prior distributions for the catchability coefficients for the trawl survey data. Martin Dorn, Jeff Grout, an anonymous reviewer, and Ana Parma are also thanked for helpful comments on the manuscript.

References

Arnold, S.F. 1990. Mathematical statistics. Prentice Hall Inc., Englewood Cliffs, N.J.

Bence, J.R., Gordoa, A., and Hightower, J.E. 1993. Influence of age-selective surveys on stock synthesis assessments. Can. J. Fish. Aquat. Sci. **50**: 827–840.

Berger, J.O. 1985. Statistical decision theory and Bayesian analysis. 2nd ed. Springer-Verlag, New York.

Box, G.E.P., and Tiao, G.C. 1973. Bayesian inference in statistical analysis. Addison-Wesley, Reading, Mass.

Clark, W.G. 1991. Groundfish exploitation rates based on life history parameters. Can. J. Fish. Aquat. Sci. **48**: 734–750.

Engås, A., and Ona, E. 1990. Day and night fish distribution pattern in the net mouth area of the Norwegian bottom-sampling trawl. Rapp. P.-V. Reun. Cons. Int. Explor. Mer, **189**: 123–127.

Francis, R.I.C.C. 1992. Use of risk analysis to assess fishery management strategies: a case study using orange roughy (*Hoplostethus atlanticus*) on the Chatham Rise, New Zealand. Can. J. Fish. Aquat. Sci. **49**: 922–930.

Francis, R.I.C.C., Robertson, D.A., Clark, M.R., and Coburn, R.P. 1992. Assessment of the QMA 3B orange roughy fishery for the 1992/93 season. N.Z. Fish. Assess. Res. Doc. No. 92/4.

Gelfand, A.E., and Smith, A.F.M. 1990. Sampling-based approaches to calculating marginal densities. J. Am. Stat. Assoc. **85**: 398–409.

Geweke, J.F. 1989. Bayesian inference in econometric models using Monte Carlo integration. Econometrica, **57**: 1317–1340.

Givens, G.H. 1993*a*. Contribution to discussion on the meeting on 'The Gibbs sampler and other Markov chain Monte Carlo methods.' J. R. Stat. Soc. B, **55**: 75–76.

Givens, G.H. 1993*b*. A Bayesian framework and importance sampling methods for synthesizing multiple sources of evidence and uncertainties linked by a complex mechanistic model. Ph.D. thesis, Department of Statistics, University of Washington, Seattle, Wash.

Gunderson, D.R. 1993. Surveys of fisheries resources. John Wiley & Sons, Inc., New York.

Harden-Jones, F.R., Margetts, A.R., Greer Walker, M., and Arnold, G.P. 1977. The efficiency of the Granton otter trawl determined by sector-scanning sonar and acoustic transponding tags. Rapp. P.-V. Reun. Cons. Int. Explor. Mer, **170**: 45–51.

Hastings, W.K. 1970. Monte Carlo sampling methods using Markov chains and their applications. Biometrika, **57**: 97–109.

Hilborn, R., and Walters, C.J. 1992. Quantitative fisheries stock assessment: choice, dynamics, and uncertainty. Chapman and Hall, New York.

Hoff, R.Z. 1989. Spatial distributions and abundance trends for two species of flatfish in the eastern Bering Sea. M.S. thesis, University of Washington, Seattle, Wash.

Ianelli, J., and Heifetz, J. 1995. Decision analysis of alternative harvest policies for the Gulf of Alaska Pacific ocean perch fishery. Fish. Res. (Amsterdam), **24**: 35–63.

Kimura, D.K. and Lyons, J.J. 1991. Between-reader bias and variability in the age-determination process. Fish. Bull. **89**: 53–60.

Kinas, P.G. 1993. Bayesian statistics for fishery stock assessment and management. Ph.D. thesis, Department of Statistics, University of British Columbia, Vancouver, B.C.

Kinas, P.G. 1996. Bayesian fishery stock assessment and decision making using adaptive importance sampling. Can. J. Fish. Aquat. Sci. **53**: 414 423.

McAllister, M.K. 1995. Using decision analysis to choose a design for surveying fisheries resources. Ph.D. thesis, School of Fisheries, University of Washington, Seattle, Wash.

© 1997 NRC Canada

McAllister, M.K., and Pikitch, E.K. 1997. A Bayesian approach to choosing a design for surveying fishery resources: application to the eastern Bering Sea trawl survey. Can. J. Fish. Aquat. Sci. **54**: 301–311.

McAllister, M.K., Pikitch, E.K., Punt, A.E., and Hilborn, R. 1994. A Bayesian approach to stock assessment and harvest decisions using the sampling/importance resampling algorithm. Can. J. Fish. Aquat. Sci. **51**: 2673–2687.

Methot, R.D. 1990. Synthesis model: an adaptable framework for analysis of diverse stock assessment data. *In* Proceedings of the Symposium on Applications of Stock Assessment Techniques to Gadids. *Edited by* L. Low. Int. North Pac. Fish. Comm. Bull. **50**: 259–277.

Metropolis, N., Rosenbluth, A.W., Rosenbluth, M.N., Teller, A.H., and Teller, E. 1953. Equations of state calculations by fast computing machines. J. Chem. Phys. **21**: 1087–1092.

Myers, R.A., Bridson, J., and Barrowman, N.J. 1993. Summary of worldwide stock and recruitment data. Can. Tech. Rep. Fish. Aquat. Sci. No. 2024.

Nash, J.C. 1990. Compact numerical methods for computers: linear algebra and function minimization. 2nd ed. Adam Higler Ltd., New York.

Newton, M.A., and Raftery, A.E. 1994. Approximate Bayesian inference with the weighted likelihood bootstrap. J. R. Stat. Soc. B, **56**: 3–48.

Oh, M.S., and Berger, J.O. 1992. Adaptive importance sampling in Monte Carlo integration. J. Stat. Comput. Simul. **41**: 143–168.

Pennington, M., and Vølstad, J.H. 1994. Assessing the effect of intra-haul correlation and variable density on estimates of population characteristics from marine surveys. Biometrics, **50**: 725–732.

Press, W.H., Teukolsky, S.A., Vetterling, W.T., and Flannery B.P. 1992. Numerical recipes in Fortran. The art of scientific computing. 2nd ed. Cambridge University Press, New York.

Punt, A.E. 1993. The implications of some multiple stock hypotheses for Chatham Rise orange roughy. N.Z. Fish. Assess. Res. Doc. No. 93/16.

Punt, A.E., Butterworth, D.S., and Penny, A.J. 1993. Stock assessment and risk analysis for the South Atlantic population of albacore (*Thunnus alalunga*). ICCAT Doc. No. SCRS/93/83. International Commission for the Conservation of Atlantic Tunas, La Jolla, Calif.

Raftery, A.E., and Lewis, S.M. 1992. How many iterations in the Gibbs sampler? *In* Bayesian Statistics 4: Proceedings of the Fourth Valencia International Meeting, April 15–20, 1991. *Edited by* J.M. Bernardo, J.O. Berger, P.A. David, and A.M. Smith. Clarendon Press, Oxford. pp. 763–773.

Raftery, A.E., Givens, G.H., and Zeh, J.E. 1995. Inference from a deterministic population dynamics model for bowhead whales. J. Am. Stat. Assoc. **90**: 402–416.

Ripley, B.D. 1987. Stochastic simulation. John Wiley & Sons, Inc., New York.

Rubin, D.B. 1987. Comment on 'The calculation of posterior distributions by data augmentation.' J. Am. Stat. Assoc. **82**: 543–546.

Rubin, D.B. 1988. Using the SIR algorithm to simulate posterior distributions. *In* Bayesian Statistics 3: Proceedings of the Third Valencia International Meeting, June 1–5, 1987. *Edited by* J.M. Bernardo, M.H. Degroot, D.V. Lindley, and A.M. Smith. Clarendon Press, Oxford. pp. 385–402.

Schnute, J.T., and Richards, L.J. 1995. The influence of error on population estimates from catch–age models. Can. J. Fish. Aquat. Sci. **52**: 2063–2077.

Smith, A.F.M. 1991. Bayesian computational methods. Philos. Trans. R. Soc. Lond. A Math. Phys. Sci. **337**: 369–386.

Stocker, M., Hand, C.M., and McAllister, M.K. 1994. Pacific cod. *In* Groundfish stock assessment for the west coast of Canada in 1993 and recommended yield options for 1994. *Edited by* M. Stocker. Can. Tech. Rep. Fish. Aquat. Sci. No. 1975. pp. 50–92.

van Dijk, H.K., and Kloek, T. 1983. Monte Carlo analysis of skew posterior distributions: an illustrative economic example. Statistician, **32**: 216–233.

Walters, C.J., and Ludwig, D. 1994. Calculation of Bayes posterior probability distributions for key population parameters: a simplified approach. Can. J. Fish. Aquat. Sci. **51**: 713–722.

Walters, C.J., and Punt, A.E. 1994. Placing odds on sustainable catch using virtual population analysis and survey data. Can. J. Fish. Aquat. Sci. **51**: 946–958.

West, M. 1992. Modeling with mixtures. *In* Bayesian Statistics 4: Proceedings of the Fourth Valencia International Meeting, April 15–20, 1991. *Edited by* J.M. Bernardo, J.O. Berger, P.A. David, and A.M. Smith. Clarendon Press, Oxford. pp. 503–524.

West, M. 1993. Approximating posterior distributions with mixtures. J. R. Stat. Soc. B, **55**: 409–422.

Wilderbuer, T.K., Walters, G.E., and Bakala, R.G. 1992. Yellowfin sole, *Pleuronectes asper*, of the eastern Bering Sea: biological characteristics, history of exploitation, and management. Mar. Fish. Rev. **54**: 1–18.

Appendix 1

Derivation of the importance function when some variables in the multivariate *t* distribution are log transformed

Some of the population dynamics model parameters (e.g., M, B_0, σ_r, q_i, s^i_{50} in Table 2) were log transformed for use in the multivariate t importance function. This was to reproduce the skewness in the marginal posteriors for these parameters and improve the efficiency of SIR (van Dijk and Kloek 1983). During importance sampling, the importance ratios, $w(\theta_k)$, are computed:

$$(1.1) \qquad w(\theta_k) = \frac{L(\text{data}\,|\,\theta_k)p(\theta_k)}{h(\theta_k)}$$

where θ_k is one particular realization of the vector of population dynamics model input parameters θ, $w(\theta_k)$ is the importance ratio, $L(\text{data}\,|\,\theta_k)$ is the likelihood function of the data evaluated at θ_k, $p(\theta_k)$ is the prior pdf evaluated at θ_k, and $h(\theta_k)$ is the importance function evaluated at θ_k (Berger 1985, p. 263). The posterior of interest is for the vector of untransformed parameters, θ. The importance function, $h(\theta)$ in eq. 1.1, thus must be a density function of the untransformed variables (van Dijk and Kloek 1983).

With log transformation of some of the variables in the multivariate t distribution, the derivation of the resulting density function for the untransformed variables is fairly simple. It can be shown that if some of the variables in a multivariate t distribution are on a log scale, then the importance function for the untransformed variables is given by

$$(1.2) \qquad h(\theta\,|\,d,\mu,V) = \text{abs}[\det J]\frac{\Gamma((d+p)/2)}{(\det V)^{1/2}(\pi d)^{p/2}\Gamma(d/2)} \times \left(1 + \frac{1}{d}(\theta'' - \mu)'V^{-1}(\theta'' - \mu)\right)^{-(d+p)/2}$$

where θ'' is the vector of estimated model input parameters, some of them log transformed, d is the degrees of freedom, μ is the mean of the vector θ'', V is a $p \times p$ positive definite matrix, and abs[det J] is the absolute value of the Jacobian,

© 1997 NRC Canada

the determinant of the matrix J whose (i,j)th component is (Arnold 1990)

$$J_{i,j} = \frac{\partial}{\partial \theta^j} z(\theta^i)$$

where $z(\theta^i)$ is the transformed parameter $\theta^{i\prime\prime}$ expressed as a function of the ith parameter θ^i.

When some of the members of the vector $\theta_k{}''$ are log transformed, J is a diagonal matrix with "1" for each of the untransformed parameters in $\theta_k{}''$ and "$1/\theta_k^j$" for each of the log transformed parameters θ_k^j where j is the index for parameter j. It can be shown that abs[det J] is simply the absolute value of the product of the terms in the diagonal of J:

$$\text{(1.3)} \qquad \text{abs}[\det J] = \text{abs}\left[\prod_{j=1}^{p}\left(\frac{1}{\theta_k^j}\right)^{(1-l_j)}\right]$$

where θ_k^j is the untransformed value for the jth parameter of the vector θ_k, $l_j = 0$ if θ_k^j is log transformed in $\theta_k{}''$ and $l_j = 1$ if not.

Note that the result in eq. 1.2 is equivalent to using an importance ratio in the transformed parameters. For example, the prior in the transformed parameters is given by

$$\text{(1.4)} \qquad p(\theta'') = \frac{p(\theta)}{\text{abs}[\det J]}.$$

This follows from the standard formula for a change of variables (Arnold 1990).

In the SIR algorithm, values for the vector θ'', some of them on a log scale, are drawn from a multivariate t distribution. This is equivalent to drawing θ_k from the density function in eq. 1.2. The density of the importance function $h(\theta)$ is evaluated at θ_k using eq. 1.2. The calculated density is then used to compute the importance ratio $w(\theta_k)$ (eq. 1.1).

Appendix 2

Likelihood function for catch-age data

The numbers of fish at age were considered to be multinomially distributed. An appropriate likelihood function for these data could be

$$\text{(2.1)} \qquad \lambda^i = \sum_{j=1}^{n_i}\sum_{a=1}^{m'} x_{a,j}^i \log(c_{a,j}^i)$$

where $x_{a,j}^i$ is the observed number of fish, $c_{a,j}^i$ is the predicted proportion of fish at age a in the jth year of series i, n_i is the number of years in series i, and m' is the plus group for the age data (18 years for both series). However, owing to the sampling methodology, the assumption of multinomial sampling does not hold. The effective sample size per year, i.e., the number of independent sample units, is believed to be much smaller than the actual number of fish aged per year. This is because the fish are not sampled one by one at random from the entire population. Instead the fish tend to range about in groups of similar ages. Thus, when they are sampled, they are scooped up in nets along with fish of similar ages. It is therefore common practice not to use the original number aged as the effective sample size. Instead, the proportions at age from the original sample are calculated by

$$\text{(2.2)} \qquad o_{a,j}^i = \frac{x_{a,j}^i}{T_{i,j}}$$

where $T_{i,j}$ is number of fish aged in year j and series i, and a much smaller number is used for the sample size in each year ($T'_{i,j} < T_{i,j}$). It is also common practice to assume that the sample size is the same in each year (e.g., Methot 1990). Hence, T_i' is used instead of $T'_{i,j}$. The effective number of fish observed in each age a, year j, and series i is given by

$$\text{(2.3)} \qquad x_{a,j}^i = T_i' o_{a,j}^i.$$

Therefore, the contribution of series i to the log likelihood is

$$\text{(2.4)} \qquad \lambda^i = \hat{T}_i \sum_{j=1}^{n_i}\sum_{a=1}^{m'} o_{a,j}^i \log(c_{a,j}^i)$$

where $\hat{T}_i$ is the estimated effective sample size for the total number of fish sampled in each year. T_i was estimated by starting with a first guess at T_i and then estimating T_i by using (see below for derivation)

$$\text{(2.5)} \qquad \hat{T}_{i,j} = \frac{\sum_{a=1}^{m'} c_{a,j}^i(1-c_{a,j}^i)}{\sum_{a=1}^{m'}(o_{a,j}^i - c_{a,j}^i)^2}.$$

The values of $c_{a,j}^i$ that were used were obtained from the modal estimate from the posterior pdf of the model input parameters. The estimated effective sample size for series i was obtained from the mean of the estimates of effective sample size over the series

$$\text{(2.6)} \qquad \hat{T}^i = \frac{1}{n_i}\sum_{j=1}^{n_i} \hat{T}_{i,j}.$$

The estimate, $\hat{T}_i$, was estimated iteratively by inputting the last estimate of $\hat{T}_i$ into the estimation procedure to obtain the mode of the posterior of model input parameters and then reestimating $\hat{T}_i$. The estimates of $\hat{T}_i$ converged at 104 and 102 for the trawl survey and catch age series, respectively.

Equation 2.5 is derived from the following:

$$\text{(2.7)} \qquad \text{var}(o_{a,j}^i) = E(o_{a,j}^i - \bar{c}_{a,j}^i)^2 = \frac{\bar{c}_{a,j}^i(1-\bar{c}_{a,j}^i)}{T_{i,j}}$$

where $\bar{c}_{a,j}^i$ is the true proportion of fish at age in series i in year j, assuming that the sample from the population is multinomially distributed. Using a normal approximation and eq. 2.7, the variance in the observed proportion can be approximated by

$$\text{(2.8)} \qquad \text{var}(o_{a,j}^i) \cong (o_{a,j}^i - c_{a,j}^i)^2 \cong \frac{c_{a,j}^i(1-c_{a,j}^i)}{T_{i,j}}$$

where $c_{a,j}^i$ is the posterior modal estimate of $c_{a,j}^i$. The estimate

© 1997 NRC Canada

of $T_{i,j}$ in eq. 2.5 is obtained by rearranging the following equality:

$$(2.9) \quad \sum_{a=1}^{m'} \mathrm{var}(o^i_{a,j}) \cong \sum_{a=1}^{m'} (o^i_{a,j} - c^i_{a,j})^2 \cong \sum_{a=1}^{m'} \frac{c^i_{a,j}(1 - c^i_{a,j})}{T_{i,j}}.$$

Appendix 3

Constructing priors for catchability in trawl survey biomass indices

The catchability coefficients, q_i, relate the trawl survey abundance indices to the biomass of yellowfin sole in the eastern Bering Sea that is potentially vulnerable to trawl survey gear. Separate prior distributions for q_i were constructed for the two trawl series, one from 1975 to 1981 and the other from 1982 to 1994. This was because the trawl survey gear was modified in 1982 to make it more efficient for bottom dwelling species (e.g., starting in 1982, two extra 61-cm sections of chain were attached to the footrope of the net so that better bottom contact was achieved, Gunderson 1993).

The method used to construct the priors for q_i is analogous to the Monte Carlo approach used by McAllister et al. (1994) to construct a prior for a constant of proportionality for an acoustic biomass series. The procedure has five steps: (*i*) identify the variables that affect q_i, (*ii*) construct q_i as an explicit function of these variables, (*iii*) construct pdfs for these variables using the scientific literature, data that are not in the likelihood function in Bayesian posterior (Equation 1), and by consulting technical experts on the processes of interest, (*iv*) estimate a distribution for q_i using Monte Carlo simulation, and (*v*) fit a parametric density function to the estimated distribution.

The mean size of q_i for each time series depends on several factors. If values for each of these factors are assumed to be independent and are randomly generated from their respective distributions, the value for the catchability coefficient for the trawl series from 1982 to 1994 can be calculated using the equation

$$(3.1) \quad q_2 = p \cdot m'' \cdot c_2 \cdot h.$$

The value for q_1 can be calculated using

$$(3.2) \quad q_1 = p \cdot m'' \cdot \frac{c_2}{r} \cdot h.$$

These factors and their distributions are explained below.

Herding (h) is the ratio of the total number of fish that encounter the net to the number that encounter the net that are immediately before the net as it approaches. This factor reflects the bias introduced by having fish herded by the trawl doors into the estimated path swept by the net. The estimated path swept in the eastern Bering Sea trawl survey is that between the wing tips of the net. The trawl doors are large metal plates that drag along the bottom and help to keep the net spread wide open. Several studies indicate that these increase the catching power of the net by scaring fish from outside into the path swept by the net (Harden Jones et al. 1977). David Somerton (AFSC, NMFS, Seattle, Wash.) has conducted some trawl experiments with trawl gear similar to that used in the eastern Bering Sea trawl survey to estimate this herding component. His studies and other studies that he is familiar with suggest the following distribution for h for flatfishes: h is a truncated Normal(2.25, 0.50^2) with $h > 0$.

Efficiency of the trawl net given that fish have encountered the net (c_i) for each trawl series i: this is the proportion of the fish that encounter the net that go into it and do not escape. Craig Rose (AFSC, NMFS, Seattle, Wash.) has conducted some underwater video experiments using the same trawl gear as that used in the eastern Bering Sea trawl survey and estimated that very few flatfish that encounter the net escape. This suggests the following distribution for c for the net used between 1982 and 1994: c_2 is a truncated Normal(0.90, 0.05^2) with $c_2 < 1$. Studies that compare the relative efficiency of the more recent gear with that of the gear used before 1982 suggest that the more recent gear may be two or more times more efficient than the older gear (Hoff 1989). The assumed distribution for this ratio, r, for several ranges of sizes of yellowfin sole in Hoff (1989) suggests the following distribution for r : r is lognormal(2.5, 0.35^2). The mean and the variance used are calculated from the means of the estimates and their variances in Hoff (1989) for size groups 20 cm and larger. The value of c_1 for the series before 1982 is given by c_2/r.

Proportion of the population of yellowfin sole in the eastern Bering Sea that on average across years is within the area of the survey (p): this variable is modeled as a function of the proportion of the biomass that is to the north of the design, P_N, and the proportion of the biomass that is to the east of the design, P_E. Work in McAllister (1995, Chap. 1) suggests that these proportions can be modeled as follows: $P_N \sim$ Uniform(0.05, 0.20) and $P_E \sim$ Uniform(0.05, 0.20). The proportion of the biomass within the design is given by

$$(3.3) \quad p = \frac{1}{1 + P_N + P_E}.$$

Bias from migration of fish within the design and vessel avoidance (Engås and Ona 1990) (m''): work in McAllister (1995, Chap. 2) suggests that if yellowfin sole migrate inshore at a rate of about 0.1 m/s, the expected bias is about −30%. At a migration rate of 0.2 m/s, this becomes −50%. At 0.5 m/s, this would be −75%. Vessel avoidance would also increase the negative bias in trawl biomass estimates. m'' was assumed to have a truncated Normal(0.60, 0.20^2) distribution with $m'' > 0$. The variance is a reflection of the amount of uncertainty in the interannual mean value of this quantity.

Any values for q_i that were less than 0.05 were discarded. Lognormal distributions were fitted to the resulting distributions for q_1 and q_2. The resulting distributions were $q_1 \sim$ lognormal(0.44, 0.59^2) and $q_2 \sim$ lognormal(0.97, 0.51^2).

© 1997 NRC Canada

A Bayesian approach to choosing a design for surveying fishery resources: application to the eastern Bering Sea trawl survey

Murdoch K. McAllister and Ellen K. Pikitch

Abstract: Decision analytic approaches (DAAs) are useful for designing surveys of fishery resources because they enable evaluation of the trade-offs of each alternative and account for uncertainty. However, such problems may require models with many uncertain parameters. We present a DAA that uses a Bayesian posterior probability density function (pdf) of multiple uncertain population model parameters. The pdf is estimated in a companion paper (this issue). Draws are taken from this pdf to evaluate the management consequences of alternative survey designs. We illustrate the approach using yellowfin sole (*Limanda aspera*) in the eastern Bering Sea. The example illustrates the bias–variance trade-off of treating trawl survey abundance estimates as relative versus absolute biomass indices. Treating the indices as absolute resulted in low variances but large biases in allowable biological catch (i.e., coefficients of variation (CVs) ≤19% and biases >50%) in all survey designs considered. Treating the indices as relative resulted in decreased absolute biases (i.e., to ≤16%) but increased variance especially for a survey once every 3 years (e.g., the CV increased from 17 to 36%).

Résumé : Les approches fondés sur les analyses de décision (AAD) sont utiles pour la conception des plans de relevé des ressources halieutiques parce qu'elles permettent une évaluation des arbitrages de chacune des choix et tient compte de l'incertitude. Toutefois, de tels problèmes peuvent exiger des modèles avec plusieurs paramètres incertains. Nous présentons une AAD qui fait appel à une densité de probabilité (dp) postérieure bayésienne des paramètres du modèle multivarié de population avec incertitude. La dp est estimée dans une communication d'accompagnement (ce numéro). Des tirages sont faits de cette dp pour évaluer les conséquences sur la gestion des différents options de plan de relevé. Nous illustrons cette approche en utilisant pour exemple la limande à nageoires jaunes (*Limanda aspera*) dans l'est de la mer de Bering. L'exemple illustre les arbitrages biais–variance lorsqu'on traite les estimations d'abondance obtenues à l'aide de relevés par chalutage comme des indices absolus ou relatifs de la biomasse. Le fait de les traiter comme des indices absolus a entraîné des variances faibles, mais des erreurs importantes dans les prises biologiques admissibles (c.-à-d. coefficients de variation (CV) ≤19% et biais >50%) pour tous les plans de relevés envisagés. Le fait de les traiter comme des indices relatifs a entraîné une diminution de l'erreur absolue (c.-à-d. ≤16%), mais une augmentation de la variance, particulièrement pour les relevés réalisés une fois tous les 3 ans (p. ex. le CV est passé de 17 à 36%). [Traduit par la Rédaction]

Introduction

Fishery managers are often faced with the task of choosing among alternative research programs that entail differing benefits and expenditures. Further research can potentially improve the estimation of quantities used in management (e.g., allowable biological catches (ABCs)) and the economic yield from the fishery. However, the benefits of conducting additional research are often highly uncertain. Decision analytic procedures that have been developed to deal with such problems (Raiffa 1968; Berger 1985) entail the following general steps. (*i*) Define the decision-makers' objectives. (*ii*) Identify the alternative actions that can be taken. (*iii*) Identify alternative hypotheses, or states of nature, that describe the uncertainty in the events or processes of interest. These hypotheses can be in the form of alternative values for the input parameters in a population dynamics model (e.g., natural mortality rate and stock–recruit parameters). (*iv*) Compute a probability distribution for the alternative states of nature. (*v*) For each state of nature, compute the potential consequences of each action. (*vi*) Summarize the results in the form of probability distributions and expected values for the consequences of each of the actions that can be taken. (*vii*) Rank alternative management actions. (*viii*) Conduct a sensitivity analysis: do the rankings change by altering certain assumptions?

A key step in decision analysis is the calculation of the probability distribution for the alternative states of nature or model input parameters. A Bayesian statistical approach is typically required for this (Berger 1985). In management problems such as the one outlined above, there are often multiple uncertain model parameters. However, applications of Bayesian decision analysis to such problems have been constrained by the absence of flexible methods to deal with multiple uncertain parameters. For example, several applications have considered relatively few (e.g., 5–10) hypotheses

Received January 23, 1996. Accepted July 31, 1996.
J13267

M.K. McAllister[1] **and E.K. Pikitch.** Fisheries Research Institute, Box 357980, University of Washington, Seattle, WA 98195-7980, U.S.A.

[1] Present address: Renewable Resources Assessment Group, Imperial College, 8 Princes Gardens, London, SW7 1NA, U.K. e-mail: mmcal@ic.ac.uk

© 1997 NRC Canada

about alternative states of nature, with each hypothesis consisting of a fixed set of values for model parameters (e.g., Sainsbury 1988; Walters and Collie 1989; Collie and Walters 1991). Such simplifications, i.e., treating uncertainty over values for model parameters in strictly discrete rather than continuous terms, can potentially bias decision analysis results (e.g., the expected economic yield of alternative policies). In contrast, Bergh and Butterworth (1987) provided a Monte Carlo procedure that estimates a continuous posterior probability distribution for eight population model parameters. However, the procedure cannot be easily generalized to other situations.

Recently, the sampling – importance resampling (SIR) algorithm (Rubin 1987, 1988) has been used to estimate Bayesian posterior probability density functions (pdfs) of population model parameters (e.g., Francis et al. 1992; Punt 1993; McAllister et al. 1994; Stocker et al. 1994; Raftery et al. 1995; McAllister and Ianelli 1997). The approach is flexible and permits estimation of a continuous posterior distribution of multiple (e.g., >50) uncertain parameters.

In this paper, we present a Bayesian decision analytic approach (DAA) to evaluate the consequences of alternative designs for surveying fishery resources. The approach employs the SIR algorithm of McAllister and Ianelli (1997) to obtain a set of draws from a Bayesian pdf of population model parameters. We illustrate the approach using yellowfin sole (*Limanda aspera*) in the eastern Bering Sea for which data on relative abundance and age frequency in the commercial catch and a trawl survey are available. The posterior distribution that is estimated in McAllister and Ianelli (1997) is used in a decision analytic framework to evaluate trade-offs among alternative trawl survey designs for yellowfin sole in the eastern Bering Sea. The example illustrates the potential impacts of conducting trawl surveys every year versus once every 3 years on the estimation of quantities used in the management of the resource (e.g., stock biomass) and on the economic yield of the fishery. The example also illustrates the bias–variance trade-off from estimating survey catchability (q) in future assessments versus fixing q at 1 (i.e., if q is estimated, biases in estimates decrease and variances increase, and vice versa if q is fixed). The example is fairly complicated but helps to illustrate that decision analysis results can be very sensitive to the details of implementation (e.g., assumptions about the relationship between future harvesting rules and the actual catches taken).

Decision analytic framework for the choice of a research survey

Under the Bayesian DAA the most desirable policy is typically the one that gives the highest expected value of some desired quantity or outcome j across all potential states of nature θ (Raiffa 1968; Berger 1985; Sainsbury 1988). For example, outcome j could be the average annual catch biomass, policy p could be a survey every year with high precision, and state of nature θ_i could be one particular realization of the set of input parameters to a population dynamics model. The state of nature θ and each outcome j of some policy p are each considered to be random variables.

Often there is a set of outcomes that is of interest, I_p, for each policy p and the decision problem is one of selecting the policy that has the least objectionable set of expected outcomes overall. In some DAAs, the various outcomes are combined into a single utility or loss function (Keeney and Raiffa 1976; Kinas 1993). However, we kept the various outcomes separate so that decision-makers can more clearly view the trade-offs among the alternatives (McAllister et al. 1994).

The expected value of outcome j, for policy p is summarized by the posterior expectation, $E^{P(\theta|\text{data})}[I_{j,p}(\theta)]$. The posterior expectation $E^{P(\theta|\text{data})}[I_{j,p}(\theta)]$ is computed using the following assumptions. Each possible outcome j, of policy p, $I_{j,p}(\theta)$, is considered to be a function of the state of nature θ and the policy p:

$$(1) \qquad I_{j,p}(\theta) = f(\theta, p)$$

and uncertainty in $I_{j,p}(\theta)$ can arise from uncertainty over θ and the success in implementation of policy p (see Sainsbury 1988). In the current paper, this latter type is represented very simplistically, i.e., by uncertainty over the relationship between future estimates of ABC and the actual amount caught. The uncertainty in θ can be expressed by a posterior pdf of θ, $P(\theta \mid \text{data})$. $P(\theta \mid \text{data})$ can be obtained by a Bayesian stock assessment that first constructs a prior pdf of θ, $p(\theta)$. The prior reflects the credibility of alternative states of nature prior to obtaining a set of data that can further our ability to discriminate among alternative θs. The population model is then fit to the data and the prior probability $p(\theta_i)$ for a given realization of θ is updated to the posterior probability through Bayes theorem:

$$(2) \qquad P(\theta_i \mid \text{data}) = \frac{L(\text{data} \mid \theta_i) p(\theta_i) d\theta}{\int L(\text{data} \mid \theta) p(\theta) d\theta}$$

where $L(\text{data} \mid \theta_i)$ is the likelihood function of the data evaluated at θ_i. If the outcome $I_{j,p}(\theta)$ is a known function of θ, then a posterior pdf of, $I_{j,p}(\theta)$, $P(I_{j,p}(\theta) \mid \text{data})$, can be constructed from the posterior pdf of θ. The uncertainty in $I_{j,p}(\theta)$ can then be expressed by this posterior pdf of $I_{j,p}(\theta)$. The posterior expectation of $I_{j,p}(\theta)$ which can be used in decision-making can be given by

$$(3) \qquad E^{P(\theta|\text{data})}[I_{j,p}(\theta)] = \frac{\int I_{j,p}(\theta) L(\text{data} \mid \theta) p(\theta) d\theta}{\int L(\text{data} \mid \theta) p(\theta) d\theta}.$$

Decision analytic procedure

The products of a Bayesian decision analytic procedure, i.e., the posterior pdf of $I_{j,p}(\theta)$ and associated quantities, e.g., $E^{P(\theta|\text{data})}[I_{j,p}(\theta)]$, can potentially assist in choosing among alternative designs for surveying fisheries resources. Such a procedure entails the following steps.

(1) For each alternative survey design of interest, specify the types of data that are to be collected and how they are to be collected (e.g., how often and how intensely). Also, specify the statistical properties of the data that are to be collected. For example, specify a probability density function for each type of data and the relationship between sampling design and the parameters of the pdf for the data. Ideally, this relationship should have some empirical basis (see Chap. 1 in McAllister 1995). Thus, each survey design will have a specific pdf of the data that will be obtained in future surveys.

(2) Specify the harvesting policy or potential set of harvesting policies that could be used in future years. For ex-

© 1997 NRC Canada

ample, this may be a harvest rate policy or constant escapement policy.

(3) Specify the time horizon and outcomes (e.g., biological, economic, and statistical–scientific) of interest that could result from each of the alternative surveying–harvest policy options. From these, construct indices of policy performance that can be used to choose among alternative policy options. For example, these could include the expected amount of depletion of the fish population at the end of the time series and the average annual economic value of the catch that can result under each surveying option.

(4) Specify the assessment procedure that will be used in future years. For example, what population dynamics model (PDM) will be fitted to the data and what fitting procedure will be used? The assessment procedure must be specified such that it can be simulated in projections of the PDM in step 4. Because the proposed decision analytic procedure is simulation based and is subject to computational limitations, the future assessment procedure is typically some shortcut of the procedure that is actually to be used.

(5) Specify a PDM that is to represent the historic and future dynamics of the fish population of interest. For example, this can be an age-structured model that entails a set of equations specifying the birth and death processes and the vulnerability of fish to fishing gear. Each potential set of values for the input parameters of this model, θ_i, is considered to be a potential state of nature (see McAllister and Ianelli 1997).

(6) Identify the parameters of the PDM in step 5 that are to be treated as uncertain (θ) and construct prior probabilities $p(\theta)$ for them (see McAllister and Ianelli 1997).

(7) Specify likelihood functions of the data $L(\text{data}\,|\,\theta)$ that the PDM is to be fit to (see McAllister and Ianelli 1997).

(8) Estimate a posterior pdf of PDM parameters $P(\theta\,|\,\text{data})$ (see McAllister and Ianelli 1997).

(9) Take a large number of draws of the set of uncertain parameters θ from the posterior pdf of step 8. The number of draws taken should be the minimum necessary to provide acceptable precision in indicators of policy performance.

(10) For each set of parameters drawn in step 9, which specifies a given historic trajectory (θ_k), carry out a future projection to evaluate the consequences of alternative policy options. Each future projection involves applying the management policy under consideration to specify a management action (e.g., set the total allowable catch (TAC)) in each year for a specific time frame, e.g., 15 years. Performance statistics for each policy are calculated during each future projection (see McAllister et al. 1994). These statistics may be particular to each year in the sequence if, for example, the immediate consequences of the current and following year's quotas are of interest. The statistics can also be averaged across all years if the overall attributes of the policy options are of interest. The end result is a pdf of policy performance indices for each policy that can be used to construct policy performance statistics such as $E^{P(\theta\,|\,\text{data})}[I_{j,p}(\theta)]$.

Evaluating the scientific and economic yield of alternative designs of the eastern Bering Sea trawl survey

Government fiscal crisis has raised questions about the value of the trawl survey in the eastern Bering Sea to the management of its fisheries. This is a multispecies groundfish survey that is conducted annually by two survey vessels to provide estimates of stock biomass and age composition for several commercially valuable species. The survey costs about US$0.84 million to conduct each year (see Appendix M in McAllister 1995). The costs of management could be reduced by changing the survey to once every 2 or 3 years or only one survey vessel and half of the sampling stations. In the latter case, the annual cost would be US$0.44 million/year (see Appendix M in McAllister 1995). It is of interest to managers to quantify the potential effects of such changes on the estimates of quantities useful to the management of fisheries in the eastern Bering Sea and on the economic yield of these fisheries. In this example, the decision analytic procedure outlined above was applied to yellowfin sole in the eastern Bering Sea to evaluate such effects.

The exploitation history of yellowfin sole extends back to the mid-1950s and there has been a highly informative history of exploitation. In the 1960s, the population was heavily exploited by foreign trawl vessels (see Fig. 1 in McAllister and Ianelli 1997) with annual catches up to about 550 000 t (Wilderbuer et al. 1992). Since the early 1970s, fishery removals have dropped off to about 100 000 t annually and abundance appears to have recovered (Wilderbuer et al. 1992). The current U.S. domestic fishery is managed by TAC quotas that are set annually. Estimates of population biomass and ABC are provided by assessment scientists. The assessment typically uses age-structured commercial fishery and trawl data and absolute estimates of biomass also from the trawl survey. Fishery managers use the ABC and other considerations to set the TAC. The ABC is obtained from the catch equation by applying the estimate of fishing mortality rate that will reduce spawner biomass per recruit to 35% of that under unexploited conditions, $F_{35\%}$ (J. Ianelli, Alaska Fisheries Science Center (AFSC), National Marine Fisheries Service (NMFS), Seattle, Wash., personal communication; Clark 1991).

Estimating a joint posterior probability distribution of population dynamics model input parameters

McAllister and Ianelli (1997) detail the estimation of the posterior pdf of θ for yellowfin sole. Here, we provide a brief overview of the PDM and data that were used to estimate the posterior pdf of θ. The model that was used to simulate the population dynamics of yellowfin sole is detailed in McAllister and Ianelli (1997): it is age structured and assumes that annual recruitment is lognormally distributed about the Ricker stock–recruit function and that there is no autocorrelation in recruitment residuals. The model also specifies a fixed fraction of individuals that will recruit annually to the fishery, for each age-class, from the unrecruited population. The fraction of individuals at each age that become susceptible to fishing mortality (i.e., recruit) is assumed to follow an asymptotic logistic function. In each year, fishing mortality is assumed to occur after natural mortality has occurred.

The PDM was fitted to commercial catch-age data, and catch-age data and biomass estimates from the trawl surveys in the eastern Bering Sea (see Table 1 in McAllister and Ianelli (1997) for the values and their coefficients of variation (CVs)). The data and others in this section were provided by J. Ianelli and G. Walters (AFSC, NMFS, Seattle, Wash.). The biomass data are treated as indices of relative abundance with

© 1997 NRC Canada

an asymptotic selectivity function. The commercial catch-age data series extends from 1964 to 1993. That for the survey extends from 1975 to 1993 (see Appendix N in McAllister 1995). In each of these data series, the observed numbers at age in each year are assumed to be multinomially distributed. The biomass data of commercial catches extend from 1954 to 1994 (see Fig. 1 in McAllister and Ianelli 1997).

Estimating the consequences of alternative survey designs

For the decision analysis, only 500 draws from the posterior pdf of θ were made. More draws would be desirable to achieve higher precision in the estimates of policy performance. However, the length of each future year computation (over 1 min for each future year) precluded this possibility. Some preliminary computations using a shortcut for the future-year estimator of stock biomass in Appendix 1 indicated that a total of 500 draws yielded precision in indices of policy performance acceptable to fishery agency personnel (McAllister 1995; G. Stauffer, AFSC, NMFS, Seattle, Wash., personal communication). See Appendix 2 for an evaluation of the precision obtainable in decision analysis results from 500 draws from the posterior pdf of θ.

For each of the historic trajectories that were drawn from the posterior pdf of θ, a further 15-year horizon was carried out to evaluate various survey design options. Each future projection entailed applying the survey policy under consideration to estimate the ABC in each of the following 15 years (i.e., 1996–2010) (the catch for 1995 is always taken to be 100 kt).

In each simulated year the following five steps were simulated.

(1) Simulate the annual assessment of yellowfin sole (see Appendix 1). We assumed that the currently used maximum likelihood procedure (Bence et al. 1993) will be applied in future years and that the results from the Bayesian assessment in McAllister and Ianelli (1997) will be ignored. Each future assessment estimated the biomass in the year of the assessment, the harvest rate that reduces spawner biomass per recruit to 35% under equilibrium conditions ($U_{35\%}$), and the ABC, the product of the former two quantities. The estimate of $U_{35\%}$ in each future year is a function of the value for natural mortality rate (M) assumed in future-year assessments and the estimate of the selectivity function in that year. Therefore, the estimates of $U_{35\%}$ can be biased and vary interannually. The model used in the assessment procedure is age structured and assumes that harvest takes place after natural mortality. It is assumed that fishing takes a fixed fraction of the number of fish in each age group with the vulnerability at age described by an asymptotic logistic function. The value for M in future assessments is assumed to be known and is fixed at 0.12, as in recent assessments (Wilderbuer et al. 1992). The data used in future assessments include only those from the 15 years prior to the assessment year rather than the full series. This was done to reduce computing time. However, this shortcut may result in underestimates of estimation performance in future stock assessments, especially ones that estimate q. The assessment model was fitted to survey biomass data that are available up to the year prior to the current assessment year and are used as absolute indices of abundance with q fixed at 1 (as in recent AFSC assessments of yellowfin sole). The model is also fitted to age frequency data in the commercial catch and trawl survey. These are available for up to 2 years prior to the assessment year. The simulated procedure found the maximum likelihood estimate of 30 parameters: these included the mean cohort size ($\bar{R}$), the lognormal residuals of cohort sizes for ages 0 to 18+ fish in the first year in the data series ($\bar{\varepsilon}_a$), residuals for age 0 fish up to 10 years prior to the current year ($\hat{\varepsilon}_y$) (beyond this window, cohort sizes are not estimable because of the approximate 9-year age at recruitment), and two parameters each for the fishery and trawl survey selectivity ogives ($\hat{s}^f_{50}$, $\hat{a}^f_{50}$, $\hat{s}^t_{50}$, and $\hat{a}^t_{50}$,) (Appendix 1).

(2) Calculate the catch for the present year. This is not straightforward because the catch of yellowfin sole is determined by several variables (L.-L. Low and G. Stauffer, AFSC, NMFS, Seattle, Wash., personal communication). First, the TAC is set by managers as a function of the ABC and other considerations such as the 2 million t cap for the TAC of all commercial species in the eastern Bering Sea. The TAC for yellowfin sole is often set less than its ABC in order to meet this latter consideration. Second, the fishery for yellowfin sole is often closed before the TAC is caught because the allowable catches of certain bycatch species, such as Pacific halibut (*Hippoglossus stenolepis*), are reached before the end of the season. To model the catches in future years, the catch in each year for the years 1996–2000 was assumed to be either the ABC or a uniform random variable between 80 and 160 kt, whichever was lower. This catch function was used because the managers of the resource believe that catches will be similar to those observed in the last 5 years, since 1990, and may vary considerably from the ABC and TAC (see Fig. 1 in McAllister and Ianelli 1997). Because factors determining the catch will be less certain further into the future, beyond the year 2000, the upper limit of the uniform random function for catch was increased to 225 000 to approximate the maximum of 223 000 t (1988) observed in the last 10 years. Otherwise the catch function was the same as for the years between 1996 and 2000.

(3) Update the number of fish at age in the true simulated population by applying natural mortality, subtracting the catch in the fishery, and adding the number of births (McAllister and Ianelli 1997).

(4) Generate the data that are to be used in future stock assessments. Biomass estimates include the biomass estimates from the trawl survey. Biomass estimates are assumed to be lognormally distributed with a mean equal to the true stock biomass multiplied by the simulated true constant of proportionality for the trawl biomass series and a CV of 0.22 in the baseline case. The value of 0.22 for the CV was chosen on the basis of estimates of this quantity for yellowfin sole in the eastern Bering Sea trawl survey (see Chap. 1 in McAllister 1995). The estimate is based on the assumption that two different vessels will be chartered once every few years and that variability in catching power among vessels is a component of the variability in trawl biomass estimates. The other data generated were the proportions of fish at age in the survey and the fishery. It is assumed that these data are multinomial samples from the true distribution at age for these catch components and that the effective sample size in future years will be 100 fish, as suggested by estimates of effective sample size in historic years (McAllister and Ianelli

© 1997 NRC Canada

Table 1. List of policy performance indices for evaluation of alternative trawl survey designs in the eastern Bering Sea.

Policy performance index	Description
NPV	Net present value of the catch of yellowfin sole using a 5% discount rate, a net value of \$200/t in 1995 dollars (J. Terry, AFSC, NMFS, Seattle, Wash.); the annual cost of the survey is subtracted from this total in each year, y: $\text{NPV} = \sum_{y=1996}^{2010} (\text{net value}(\text{catch}_y) - \text{cost}(\text{survey}_y))e^{-(y-1995)0.05}$
CV in catch	Coefficient of variation in the catch reflecting the variability in the catch over the 15-year time horizon
RMSE (estimated biomass)	Root mean square error in the estimated versus simulated true recruited biomass, where $\text{RMSE}(X) = \left(\frac{1}{500}\sum_{i=1}^{500}\frac{1}{15}\sum_{y=1996}^{2010}\frac{(\hat{X}_y - X_y)^2}{X_y^2}\right)^{1/2}$
RMSE ($U_{35\%}$)	RMSE of the estimated versus true $U_{35\%}$ value for the policy
RMSE (ABC)	RMSE of the estimated versus the true ABC
Bias (estimated biomass)	Bias in the estimated recruited biomass relative to the true value for biomass averaged over years
Bias ($U_{35\%}$)	Bias in the estimated $U_{35\%}$ relative to the true value for $U_{35\%}$ averaged over years
Bias (ABC)	Bias in the estimated ABC relative to the true value averaged over years
$\%B_{2010}/B_0$	Ratio of the recruited biomass in the year 2010 to the unexploited biomass, expressed as a percentage

Table 2. Alternative policy scenarios evaluated for yellowfin sole in the eastern Bering Sea.

Frequency	Harvest policy	Estimate q?	CV (biomass)	Catch ~ uniform RV	Posterior
Every year	$F_{35\%}$	No	0.22	Yes	Baseline
Every third year	$F_{35\%}$	No	0.22	Yes	Baseline
Every year	$F_{35\%}$	Yes	0.22	Yes	Baseline
Every third year	$F_{35\%}$	Yes	0.22	Yes	Baseline
Every year	$F_{35\%}$	No	0.22	Catch set to ABC	Baseline
Every third year	$F_{35\%}$	No	0.22	Catch set to ABC	Baseline
Every year	$F_{35\%}$	Yes	0.22	Catch set to ABC	Baseline
Every third year	$F_{35\%}$	Yes	0.22	Catch set to ABC	Baseline
Every year	$F_{35\%}$	$q = 1.6$, $M = 0.14$	0.22	Yes	Baseline
Every third year	$F_{35\%}$	$q = 1.6$, $M = 0.14$	0.22	Yes	Baseline
Every year	$F_{35\%}$	No	0.15	Yes	Baseline
Every year	$F_{35\%}$	No	0.28	Yes	Baseline
Every year	$F_{35\%}$	Yes	0.15	Yes	Baseline
Every year	$F_{35\%}$	Yes	0.28	Yes	Baseline

1997). It is assumed that the catch biomass is known without error.

(5) Calculate and store performance statistics (Table 1) to assess the estimation performance of the various quantities such as stock biomass, the ABC, the economic performance of the fishery (e.g., the present value of future catches less the annual cost of each design), and the conservation of the population (e.g., the ratio of stock biomass in the final year to the average unexploited stock biomass, B_0).

The survey designs evaluated (Table 2) include ones in which a survey was conducted every year (annual design) and once every 3 years (triennial design). Furthermore, three levels of sampling CVs were considered for the trawl survey, 0.22 (baseline), 0.15, and 0.28. The value of 0.15 could

© 1997 NRC Canada

Table 3. Policy performance indices for alternative policy scenarios for trawl surveys in the eastern Bering Sea (the acronyms for the performance indices are defined in Table 1; the present value (PV), the NPV less costs, is shown in parentheses).

Statistic	NPV and (PV) ($millions)	%CV catch/ABC	%RMSE/bias biomass	%RMSE/bias $U_{35\%}$	%RMSE/bias ABC	$E(\%(B_{2010}/B_0))$
Fix q at 1.0 and M at 0.12 in future assessments						
Annual design (baseline)	272 (281)	23/15	86/86	19/–19	56/55	82
Triennial design (baseline)	279 (282)	23/17	91/91	19/–19	62/61	82
Survey CV = 0.15	274 (283)	23/12	88/88	19/–19	54/54	81
Survey CV = 0.28	271 (275)	23/17	85/85	19/–19	58/57	82
Catch = ABC (annual)	326 (335)	15/15	93/93	19/–19	60/60	72
Catch ABC (triennial)	338 (341)	18/18	98/98	19/–19	68/67	72
Fix q at 1.6 and M at 0.14 in future assessments						
Annual design	228 (237)	16/15	13/–2	8/–2	17/0	93
Triennial design	235 (239)	19/19	16/–1	8/–2	23/4	94
Estimate q and fix M at 0.12 in future assessments						
Annual design	198 (207)	23/28	36/–35	15/–15	33/–16	106
Triennial design	210 (213)	28/36	35/–33	16/–16	36/–9	105
Survey CV = 0.15	200 (209)	20/24	32/–32	15/–15	30/–16	105
Survey CV = 0.28	201 (205)	24/28	35/–34	15/–15	33/–16	106
Catch = ABC (annual)	209 (217)	26/26	32/–31	15/–15	31/–16	104
Catch = ABC (triennial)	227 (230)	36/36	32/–31	15/–15	37/–9	103

potentially be obtained if the same two vessels were used in each year and interannual variability in catching power among vessels was eliminated (see Chap. 1 in McAllister 1995). This contrasts with the current regime in which two different vessels can be chartered for the trawl survey once every 3 years. The value of 0.28 could be obtained if only one vessel was used in the survey, only half as many sampling stations were used in the survey, and a different survey vessel was used every year (see Chap. 1 in McAllister 1995).

Tests were conducted to assess the sensitivity of decision analysis results to (*i*) how the catch is modeled, (*ii*) the values used for q and M in future assessments, and (*iii*) estimating q in future assessments versus fixing it at one. Specifically, the tests were as follows: (*i*) set the catches at the ABC rather than assuming that catch is a uniformly distributed random variable, (*ii*) in future assessments, fix the values for q and M at their posterior expectations, i.e., 1.6 and 0.14, respectively, and (*iii*) estimate q rather than treat it as equal to 1 in future assessments.

Results

The results of the decision analysis are presented in the form of expected values and, in some cases, marginal posterior distributions for the performance indices. With q fixed at 1, the differences between the annual and triennial designs are very small for all policy performance indices. Under the annual (baseline) survey design, the expected net present value (NPV) of the fishery for yellowfin sole (using a 5% discount rate and a 15-yr time horizon, see Table 1) is very large, even when the cost of the survey is factored in (e.g., $272 million, Table 3). The NPV of the fishery increased by 2.3% when the design was changed from the annual to the triennial design. This gain is largely due to the reduced

© 1997 NRC Canada

cost of the survey, since the present value (i.e., when surveying costs are not included) remained virtually the same with the change to a triennial design (i.e., the increase was only 0.1%). The CV in the catch increased by only 1.4% by changing to a triennial design. The expected amount of depletion (i.e., $E(B_{2010}/B_0)$) decreased slightly (e.g., by 0.4%) when the design was changed from an annual to a triennial one (Table 3). The root mean square error (RMSE) and bias in estimated quantities such as B_y, $U_{35\%}$, and ABC_y increased little by changing to a triennial design (e.g., by 5, 0.4, and 9%, respectively, for both RMSE and bias). The bias in estimated biomass and ABC was positive and fairly large for the baseline and the triennial designs (e.g., the ABC was on average overestimated by 55 and 61%, respectively) (Table 3). This is because q was fixed at 1 in future assessments and is considerably less than the posterior expectation of q of 1.63. The bias in estimates of $U_{35\%}$ is negative (e.g., −19% for both designs). This is because M was fixed at 0.12 in future assessments and is less than the posterior expectation of 0.14. Note that the bias in the ABC decreased because of the opposing biases in $U_{35\%}$ and the estimated biomass.

Despite the sharp posteriors for model input parameters, the uncertainty in the policy indices is fairly large (Fig. 1). For example, the 90% Bayesian confidence interval (CI) for the NPV is \$239 million to \$305 million for the baseline policy and \$244 million to \$312 million for the triennial design. The 90% CIs for the 15-year CV in the catch for these same policies are 14–31 and 15–31%. Those for the amount of depletion at the end of the time horizon are 58–110 and 59–110%, respectively.

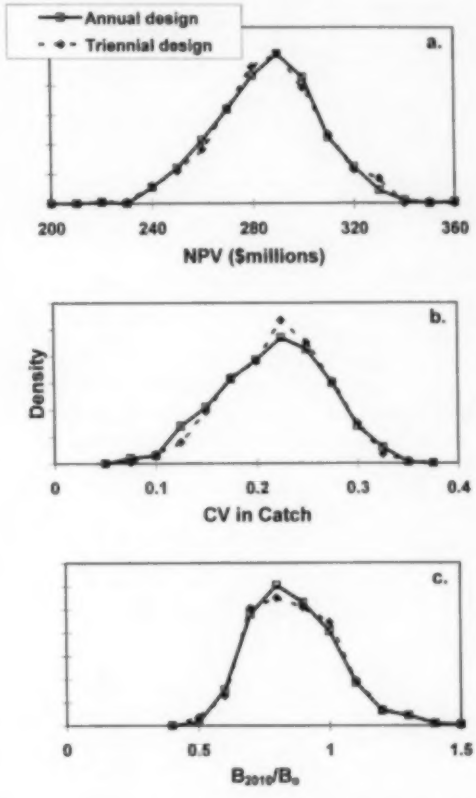
Decreasing the CV in biomass estimates to 0.15 resulted in little change in policy performance indices for the baseline design (e.g., the NPV increased by 0.7% by decreasing the CV) (Table 3). Increasing the CV to 0.28 increased the RMSE in estimated management quantities very little (e.g., by 3.6% for the estimated ABC). The NPV of the fishery decreased slightly (by 0.4%) (Table 3).

In the first sensitivity analysis, the catches were set to the ABC rather than assumed to be uniformly distributed. The NPV is considerably larger than that under the baseline design (e.g., \$326 million and \$338 million for the annual and the triennial designs, respectively) (Table 3). Estimation performance was not much different from the baseline case, as expected (e.g., the bias in the ABC was 60 and 68% for the annual and triennial designs, respectively). The depletion in stock biomass for both the annual and the triennial design decreased to 72%.

In the second sensitivity analysis, we evaluated the effect of fixing q and M in future assessments at their posterior expectations, i.e., 1.6 and 0.14. As expected, the bias in estimates of biomass, $U_{35\%}$, and ABC in future assessments and the NPV dropped considerably relative to the baseline case (Table 3). For example, the bias in the ABC decreased to 0 and 4% and the NPV decreased to \$228 million and \$235 million for the annual and triennial designs, respectively. The differences in policy performance between the annual and triennial designs increased slightly relative to the baseline case (Table 3). For example, the interannual CVs in the ABC are 15 and 19% compared with 15 and 17% in the baseline case for the annual and triennial designs, respectively.

In the third sensitivity analysis, we evaluated the effect

Fig. 1. Marginal posterior pdfs of (*a*) NPV, (*b*) the CV in catches, and (*c*) stock depletion (B_{2010}/B_0). The marginals are shown for the annual and triennial surveys.



of estimating q for the trawl survey in each future assessment. The relative difference in most performance indicators between the annual and triennial designs increased with respect to the baseline case (Table 3). The RMSE in estimates of the ABC decreased from 56 and 62% to 33 and 36%, respectively, for the annual and triennial designs when q was estimated as opposed to fixed at 1 in future assessments. This is mainly due to a large decrease in the expected bias in biomass estimates when q was estimated as opposed to fixed at 1 in future assessments (e.g., the bias in biomass estimates decreased to −35 and −33% for the triennial and annual designs, respectively). The negative biases occurred because the assumed value for M in the simulated assessments (0.12) was smaller than the posterior expectation (0.14). Furthermore, the trawl abundance indices are weighted much more heavily in the likelihood function in simulated assessments (Appendix 1) than the Bayesian one and decrease very strongly in the 1980s (see Fig. 1 in McAllister and Ianelli 1997). This caused future assessments to indicate greater depletion and smaller stock sizes than the true simulated values given by the Bayesian posterior.

The NPVs were smaller when q was estimated as opposed to fixed (e.g., \$198 million and \$210 million) because estimates of biomass no longer had strong positive bias. When q was estimated, the CV in the ABC increased from 15 and

© 1997 NRC Canada

17% to 28 and 36% for the annual and triennial designs, respectively (Table 3). The CVs in catch for the annual and triennial designs are smaller than those for the ABC (i.e., 23 and 28%) because modeled catches were uniformly distributed and less than the ABCs. When the catches were modeled to match the ABCs, the CVs in catch increased to 26 and 36% for the annual and triennial designs, respectively. The NPV increased to $209 million and $227 million, respectively, for these designs. When the CV in biomass estimates was changed, e.g., from 22 to 15%, the changes in performance indices were small, as in the case in which q was fixed at 1 in future assessments (e.g., the RMSE in ABC for CVs of 15 and 28% were 30 and 33%, respectively) (Table 3).

Discussion

Comparisons with other decision analytic studies of fisheries research surveys

Several other studies have assessed the potential trade-offs of alternative stock assessment research designs (e.g., Bergh and Butterworth 1987; Sainsbury 1988; Walters and Collie 1989; Collie and Walters 1991; Powers and Restrepo 1993). Sainsbury (1988), Walters and Collie (1989), and Collie and Walters (1991) used DAAs to evaluate alternative experimental harvesting regimes that tested alternative hypotheses about population dynamics. Unlike the current work, alternative hypotheses about population dynamics were considered in discrete rather than continuous terms (i.e., a small set of (e.g., 5–10) hypotheses were identified, each consisting of a unique realization of the values of the parameters in the models considered). This was sufficient to demonstrate the potential benefits of experimental policies. The DAA in the current work could easily be adapted to evaluate such policies. Use of a continuous joint posterior pdf of alternative states of nature could avoid potential biases from discretizing states of nature into relatively few alternatives. However, as in our example, the application of SIR would be computationally more demanding and simplification of simulated future stock assessments would be necessary.

Bergh and Butterworth (1987) applied a Bayesian-like Monte Carlo approach to evaluate the potential trade-offs between risk and economic yield of alternative harvest strategies and amounts of precision in hydroacoustic estimates of South African anchovy (*Engralis capensis*) spawning biomass and recruitment. Higher precision in survey estimates yielded larger catches for each given level of risk of obtaining a low stock size. Their DAA, however, used a Bayesian Monte Carlo procedure that is not easily generalized to instances with extensive age frequency data.

Powers and Restrepo (1993) also used a DAA to evaluate the potential net economic value of alternative research programs to enhance the precision in estimates of various PDM parameters for king mackerel (*Scomberomorus cavalla*) in the Gulf of Mexico. They considered the value of different research programs under risk-prone versus risk-averse harvesting strategies. They found that moderate increases in spending on research could yield marked gains in net economic value, especially since the stock was recovering from recent overfishing. A non-Bayesian bootstrap approach that applied a virtual population analysis based estimation procedure to catch-age and survey biomass data was used to quantify uncertainty in model input parameters. Walters and Ludwig (1994) discussed the relative merits of Bayesian and non-Bayesian approaches to fisheries risk assessment.

Powers and Restrepo (1993) also used a retrospective approach: the effects on "this" year's and "next" year's management of different research regimes were evaluated under the assumption that the data currently available had been collected under each of the alternative research regimes. This approach has the advantage of requiring only a few years of future projections to evaluate the impact of alternative research programs. The approach can, thereby, require far fewer computations and permit more Monte Carlo replicates and higher precision than the approach used in the current paper. However, unlike the approach in the current paper, Powers and Restrepo's (1993) approach does not address questions about the expected impacts of a change in research survey design in the near future.

Bias–variance trade-off

The results of our application illustrate the bias–variance trade-off from treating survey indices as absolute abundance indices rather than relative indices. Treating survey indices as absolute (i.e., fixing q at 1) in future assessments resulted in relatively low variance in estimates of quantities such as the ABC (i.e., CVs ≤ 0.19) in the designs considered. However, large positive biases (e.g., >50%) resulted. This is because the Bayesian estimate of q for the years 1982–1994 from the stock assessment was 1.6. This estimate is plausible because of recent research that suggests, for some species, that the assumed path swept by the net underestimates the effective path swept because of the effect of herding by the trawl doors (D. Somerton, AFSC, NMFS, Seattle, Wash.). In contrast, treating survey indices as relative (i.e., estimating q) in future assessments reduced the absolute bias (i.e., to $\leq$16%) but increased variance in estimates of the ABC (i.e., $0.24 \leq$ CV ≤ 0.36) in the designs considered. The results suggest that if q is estimated, a change from an annual to a triennial design may markedly lower precision in estimates of management quantities such as the ABC (e.g., the CV increased from 0.28 to 0.36) and result in poorer management performance.

Problems with NPV as an indicator of economic consequences

In contrast with other evaluations, our example suggests that NPV will not be increased and estimation performance will be increased very little by increasing sampling effort. This is for a variety of reasons. For example, the catch of yellowfin sole in recent years has regularly fallen short of the TAC and was modeled this way in the future. Therefore, catch was little affected by the research survey design. However, catch was only slightly more affected when catches were set to the ABC. Estimates of biomass and the ABC were also little affected by survey design because future assessments were modeled with q fixed at 1, as in historic assessments. Changing from an annual to a triennial design or decreasing the CV in survey biomass estimates from 0.22 to 0.15 resulted in relatively small changes in the precision in estimates of biomass and the ABC, and in NPV.

It is likely that the economic value of the fishery would be more sensitive to survey design if it were managed according to the MSY fishing mortality rate, F_{MSY}, the fishing mortality

© 1997 NRC Canada

rate that produces the maximum sustainable yield. Instead, the fishery for yellowfin sole is managed according to an $F_{35\%}$ rule that appears to be a relatively light exploitation rate (about 10% on average) for a species with such high productivity (see Fig. 3*a* in McAllister and Ianelli 1997). Under F_{MSY}, overestimates of the ABC would be more likely to result in overexploitation and underestimates in foregone economic yield. Evaluation of such an option would be computationally more demanding because the parameters for a stock–recruit relationship would also have to be estimated in each future year. It would also be prudent to apply the same decision analytic procedure to other commercially important species in the survey before any decisions were made to modify the survey.

The NPV is also relatively insensitive to survey design because only yellowfin sole is included in the evaluation. In contrast, the eastern Bering Sea trawl survey is a multispecies trawl survey and there are several other commercially valuable species surveyed. When only the economic value of the catch of yellowfin sole is considered (i.e., surveying costs are left out), the designs with low CV had present values over $1 million larger than the baseline design in both approaches for dealing with q in future assessments (Table 3). This suggests that when other species and the same surveying costs are included in the calculation of NPV, designs with higher precision may have higher NPV. Furthermore, the expected net economic value of the catch was larger for annual than triennial designs in the latter half of the 15-year projections for all cases evaluated (e.g., by $85 000 to $270 000/year). It would be desirable to use a longer time horizon and perhaps even lower discount rates for the economic evaluation of alternative designs. However, this was not considered because of computational limitations.

While the economic value of the yellowfin sole fishery was relatively insensitive to alternative survey designs, this may not be so for other species in the eastern Bering Sea. For example, the catches of walleye pollock (*Chalcogramma theragra*) tend to closely follow the recommended ABCs. This is because the TAC for walleye pollock is seldom reduced when TAC adjustments are made to meet the 2 million t cumulative TAC for the eastern Bering Sea. Furthermore, the fishery for it is seldom closed down because of bycatch.

Lastly, other issues that are not considered in the current decision analysis also warrant consideration when decisions are made about altering the existing design. For example, sudden shifts in oceanographic regimes can occur in the northeast Pacific Ocean that may affect the production of various fish populations (Hare and Francis 1995). Abrupt changes in population dynamics may be difficult to detect with imprecise, and infrequent (e.g., triennial), surveys. Evaluation of such a scenario using the current decision analytic framework could reveal how well such changes could be detected by alternative survey designs. There is also a scientific value of having a long, uninterrupted data series on several species that has been collected using a consistent protocol from year to year. The eastern Bering Sea trawl survey has been conducted more or less consistently every year since 1982, a reasonably long series for a trawl survey. Furthermore, for several species, the sampling CV is relatively small because of the fairly high number of stations sampled and the large geographic extent of the survey (see Chap. 1 in McAllister 1995). Continuing collection and use of a fishery-independent source of data in stock assessments such as those provided by the eastern Bering Sea trawl survey lend credibility to stock assessments and to the management decisions made. Without such added credibility, management would be more difficult and prone to failure. Finally, annual data provided by such surveys are often used in scientific studies apart from the regular stock assessments (e.g., Wilderbuer et al. 1992).

Overall, the application of the DAA to yellowfin sole in the eastern Bering Sea demonstrated the following. (*i*) Lower precision and less frequent surveys do not necessarily result in lower net present value of the fishery when the costs of surveying are incorporated. This was the result for yellowfin sole in the example above because the catches were modeled to be on average less than the ABC. Furthermore, the survey is multispecies, but the analysis was for yellowfin sole only. There may also be several results that are not expressed or accounted for by NPV. For example, it may take several years before the effects of a change in survey design are fully realized (e.g., 15 years). Therefore, even at a low discount rate, long-term decreases in annual net economic yield can be easily overridden by short-term increases that result from decreased surveying costs. Therefore, NPV, despite common use (e.g., Clark et al. 1985; Sainsbury 1988; Powers and Restrepo 1993), may fail to convey important economic consequences of changes in survey design. (*ii*) When survey biomass estimates are treated as absolute estimates of biomass, the differences in estimation and management performance between annual and triennial survey designs and designs with low versus high precision can be relatively small. (*iii*) Large biases can result and uncertainties may be underestimated if scientists treat constants of proportionality (q) as known without error and survey biomass values as absolute estimates of biomass. (*iv*) When survey biomass estimates are treated as relative abundance indices, variances in estimated quantities can increase and management performance can become considerably worse under a triennial design than an annual one, especially if catches closely approximate the estimated ABCs.

Acknowledgments

This work was funded by the National Marine Fisheries Service. We thank Jim Ianelli for helpful discussions, providing fishery and survey data, and help in getting the computer software going. We also thank Tom Wilderbuer and Gary Walters for providing survey and fishery data. We thank Gary Stauffer, Ray Hilborn, Judy Zeh, Don Gunderson, Rich Marasco, and Loh-Lee Low for helpful discussions. Andre Punt is thanked for helpful discussions and comments on the manuscript. Jeff Grout, an anonymous reviewer, and Ana Parma are also thanked for comments on the manuscript. Joe Terry is thanked for providing economic data on yellowfin sole.

References

Bence, J.R., Gordoa, A., and Hightower, J.E. 1993. Influence of age-selective surveys on stock synthesis assessments. Can. J. Fish. Aquat. Sci. **50**: 827–840.

Berger, J.O. 1985. Statistical decision theory and Bayesian analysis. 2nd ed. Springer-Verlag, New York.

© 1997 NRC Canada

Bergh, M.O., and Butterworth, D.S. 1987. Towards rational harvesting of the South African anchovy considering survey imprecision and recruitment variability. S. Afr. J. Mar. Sci. **5**: 937–951.
Clark, W.G. 1991. Groundfish exploitation rates based on life history parameters. Can. J. Fish. Aquat. Sci. **48**: 734–750.
Clark, C.W., Charles, A.T., Beddington, J.R., and Mangel, M. 1985. Optimal capacity decisions in a developing fishery. Mar. Res. Econ. **2**: 25–53.
Collie, J.S., and Walters, C.J. 1991. Adaptive management of spatially replicated groundfish populations. Can. J. Fish. Aquat. Sci. **48**: 1273–1284.
Francis, R.I.C.C., Robertson, D.A., Clark, M.R. and Coburn, R.P. 1992. Assessment of the QMA 3B orange roughy fishery for the 1992/93 season. N.Z. Fish. Assess. Res. Doc. No. 92/4.
Hare, S.R., and Francis, R.C. 1995. Climate change and salmon production in the northwest Pacific Ocean. *In* Climate change and northern fish populations. *Edited by* R.J. Beamish. Can. Spec. Publ. Fish. Aquat. Sci. No. 121. pp. 357–373.
Keeney, R., and Raiffa, H. 1976. Decisions with multiple objectives. John Wiley & Sons, Inc., New York.
Kinas, P.G. 1993. Bayesian statistics for fishery stock assessment and management. Ph.D. thesis, Department of Statistics, University of British Columbia, Vancouver, B.C.
McAllister, M.K. 1995. Using decision analysis to choose a design for surveying fisheries resources. Ph.D. thesis, School of Fisheries, University of Washington, Seattle, Wash.
McAllister, M.K., and Ianelli, J.N. 1997. Bayesian stock assessment using catch–age data and the sampling – importance resampling algorithm. Can. J. Fish. Aquat. Sci. **54**: 284–300.
McAllister, M.K., Pikitch, E.K. Punt, A.E., and Hilborn, R. 1994. A Bayesian approach to stock assessment and harvest decisions using the sampling/importance resampling algorithm. Can. J. Fish. Aquat. Sci. **51**: 2673–2687.
Methot, R.D. 1990. Synthesis model: an adaptable framework for analysis of diverse stock assessment data. *In* Proceedings of the Symposium on Applications of Stock Assessment Techniques to Gadids. *Edited by* L. Low. Int. North Pac. Fish. Comm. Bull. **50**: 259–277.
Powers, J.E., and V.R. Restrepo. 1993. Evaluation of stock assessment research for Gulf of Mexico king mackerel: benefits and costs to management. N. Am. J. Fish. Manage. **13**: 15–26.
Punt, A.E. 1993. The implications of some multiple stock hypotheses for Chatham Rise orange roughy. N.Z. Fish. Assess. Res. Doc. No. 93/16.
Raftery, A.E., Givens, G.H., and Zeh, J.E. 1995. Inference from a deterministic population dynamics model for bowhead whales. J. Am. Stat. Assoc. **90**: 402–416.
Raiffa, H. 1968. Decision analysis. Introductory lectures on choices under uncertainty. Addison-Wesley, Reading, Mass.
Rubin, D.B. 1987. Comment on 'The calculation of posterior distributions by data augmentation.' J. Am. Stat. Assoc. **82**: 543–546.
Rubin, D.B. 1988. Using the SIR algorithm to simulate posterior distributions. *In* Bayesian Statistics 3: Proceedings of the Third Valencia International Meeting, June 1–5, 1987. *Edited by* J.M. Bernardo, M.H. Degroot, D.V. Lindley, and A.M. Smith. Clarendon Press, Oxford. pp. 385–402.
Sainsbury, K.J. 1988. The ecological basis of multispecies fisheries and management of a demersal fishery in tropical Australia. *In* Fish population dynamics. 2nd ed. *Edited by* J.A. Gulland. John Wiley & Sons, Inc., New York. pp. 349–382.
Stocker, M., Hand, C.M., and McAllister, M.K. 1994. Pacific cod. *In* Groundfish stock assessment for the west coast of Canada in 1993 and recommended yield options for 1994. *Edited by* M. Stocker. Can. Tech. Rep. Fish. Aquat. Sci. No. 1975. pp. 50–92.
Walters, C.J., and Collie, J.S. 1989. An experimental strategy for groundfish management in the face of large uncertainty about stock size and production. *In* Effects of ocean variability on recruitment and an evaluation of parameters used in stock assessment models. *Edited by* R.J. Beamish and J.A. McFarlane. Can. Spec. Publ. Fish. Aquat. Sci. No. 108. pp. 13–25.
Walters, C.J., and Ludwig, D. 1994. Calculation of Bayes posterior probability distributions for key population parameters: a simplified approach. Can. J. Fish. Aquat. Sci. **51**: 713–722.
Wilderbuer, T.K., Walters, G.E., and Bakala, R.G. 1992. Yellowfin sole, *Pleuronectes asper*, of the eastern Bering Sea: biological characteristics, history of exploitation, and management. Mar. Fish. Rev. **54**: 1–18.

Appendix 1

Estimation procedure used within future projections

The estimation procedure that is assumed to be used in future years is the stock synthesis procedure (Methot 1990). A highly simplified version of this procedure was simulated. This version used an age-structured PDM that assumed that the natural mortality rate is constant with age and that the fishery and survey selectivity ogives are asymptotic logistic functions, as in eqs. 8 and 18 in McAllister and Ianelli (1997).

Resource dynamics

The dynamics of animals aged 2 and above are governed by the equation

$$(1.1)\quad \begin{aligned} \hat{N}_{y+1,a} &= e^{-\hat{M}}(\hat{N}_{y,a-1}(1-\hat{\phi}^f_a\hat{H}_y)) && 2 \le a < m \\ \hat{N}_{y+1,m} &= e^{-\hat{M}}(\hat{N}_{y,m}+\hat{N}_{y,m-1})(1-\hat{\phi}^f_m\hat{H}_y)) && a = m \end{aligned}$$

where $\hat{N}_{y,a}$ is the number of animals of age a at the start of year y, $\hat{\phi}^f_a$ is the proportion of animals of age a that are vulnerable to harvest:

$$(1.2)\quad \hat{\phi}^f_a = \frac{1}{1+\exp(-\hat{s}^f_{50}(a-\hat{a}^f_{50}))},$$

$\hat{s}^f_{50}$ is the slope of the fishery recruitment ogive at 50% recruited, $\hat{a}^f_{50}$ is the age at 50% recruited, $\hat{M}$ is the instantaneous rate of natural mortality on animals (fixed at 0.12), $\hat{H}_y$ is the harvest rate during year y and occurs after natural mortality (applies to all fully selected individuals), and m is the maximum (lumped) age-class (all animals in this and the previous age-class are recruited and mature) and is assumed to be identical to that in McAllister and Ianelli (1997).

Births

No stock–recruit function is assumed in this estimation. Recruitment is assumed to be governed by the equation

$$(1.3)\quad \hat{R}_y = \bar{R}\exp(\hat{\varepsilon}_y)$$

where $\bar{R}$ is the mean cohort strength over the number of age 0 fish in years following the first year that age data are used, and over the number at age in the first year that there are data, and $\hat{\varepsilon}_y$ is the estimated lognormal recruitment residual in year y.

Initial conditions

The initial numbers at age are given by the equation

$$(1.4)\quad \hat{N}_{y_1,a} = \bar{R}e^{\bar{z}a} \qquad 1 \le a \le m$$

© 1997 NRC Canada

where $\tilde{\epsilon}_a$ is the lognormal residual for cohort size of fish at age a in the first year, y_1, in the simulated assessment.

Catches

The exploitation rate during year y, H_y, is obtained by solving the equation

$$\hat{H}_y = \frac{C_y}{\hat{B}_y}, \quad (1.5)$$

where C_y is the annual catch biomass observed during year y and $\hat{B}_y$ is the exploitable biomass in year y:

$$\hat{B}_y = e^{-\hat{M}} \sum_{a=1}^{m} w_a \hat{\phi}_a^f \hat{N}_{y,a} \quad (1.6)$$

where w_a is the mass of a fish at age a (assumed identical to that in McAllister and Ianelli 1997).

Data series

The trawl survey data are assumed to be proportional to the biomass that is susceptible to the survey gear (i.e., $\hat{B}'_y$):

$$\hat{B}'_y = e^{-\hat{M}} \sum_{a=1}^{m} \hat{\phi}_a^t (w_a \hat{N}_{y,a} (1 - 0.5 \hat{\phi}_a^f \hat{H}_y)) \quad (1.7)$$

where $\hat{\phi}_a^t$ is the proportion of individuals at age a that are vulnerable to the survey gear,

$$\hat{\phi}_a^t = \frac{1}{1 + \exp(-\hat{s}_{50}^t (a - \hat{a}_{50}^t))}, \quad (1.8)$$

$\hat{s}_{50}^t$ is the slope of the selectivity ogive at 50% selected, and $\hat{a}_{50}^t$ is the age at 50% selected. Only a 15-year window of survey biomass data was assumed to be used to save computational time for future projections. Survey biomass data were assumed to be included up to the year prior to the assessment year, the year for which an ABC was estimated.

The proportions at age in the commercial catch are assumed to be given by

$$\hat{c}_{a,y}^f = \frac{\hat{\phi}_a^f \hat{N}_{a,y}}{\sum_{a'=1}^{m} \hat{\phi}_{a'}^f \hat{N}_{a',y}}. \quad (1.9)$$

The proportions-at-age in the trawl survey are assumed to be given by

$$\hat{c}_{a,y}^t = \frac{\hat{\phi}_a^t ((1 - 0.5 \hat{\phi}_a^f \hat{H}_y) \hat{N}_{a,y})}{\sum_{a'=1}^{m} \hat{\phi}_{a'}^t (1 - 0.5 \hat{\phi}_{a'}^f \hat{H}_y) \hat{N}_{a',y})}. \quad (1.10)$$

Age data were assumed to be included up to 2 years prior to the assessment year, the year for which an ABC was estimated (e.g., if the assessment year is 1996, age data are used up to the year 1994).

Likelihood function

The same assumptions about data errors (e.g., multinomial age compositions) were used in each future assessment as in the one for the Bayesian estimation procedure in McAllister and Ianelli (1997). However, in recent assessments of yellowfin sole, considerable emphasis has been given to the biomass data as absolute abundance indices such that the biomass series predicted by the population dynamics model has been constrained to fit closely with the observed values. To achieve this effect in the simulated future assessments, the log likelihood of the survey biomass data in future assessments was multiplied by a factor of 100 and then added to the log likelihood for the proportion-at-age data.

Appendix 2

Evaluation of the precision obtainable in decision analysis results

For the decision analysis, only 500 draws from the posterior were made. More draws would be desirable to achieve higher precision in the estimates of policy performance. However, the great length of each future year computation (over 1 min for each future year) precluded this possibility. Some preliminary computations using a shortcut to the future-year estimator of stock biomass indicated that 500 draws was not unreasonable. Fifteen sets of 500 independent sequences of draws were used to compute 15 sets of decision analysis results for a few different policies. The relative difference, RD_i, in each of the policy indicators (Table 1) between two policies was computed for each of the 15 sets of draws:

$$RD_i = \frac{(\bar{I}_{i,1} - \bar{I}_{i,2})}{\bar{I}_{i,1}} \quad (2.1)$$

where $\bar{I}_{i,j}$ is the expected value for a policy performance index from replicate i and policy j. The mean, $\overline{RD}$, and CV in the relative difference were also computed across the 15 replicates:

$$\overline{RD} = \frac{1}{15} \sum_{i=1}^{15} RD_i \quad (2.2)$$

$$CV = \frac{\left(\frac{1}{14} \sum_{i=1}^{15} (RD_i - \overline{RD})^2 \right)^{1/2}}{\overline{RD}}. \quad (2.3)$$

The CV in the relative difference in policy performance between policies was no larger than 0.15 for both large and small relative differences, $\overline{RD}$, and was less than 0.10 for most quantities. Increasing the the number of draws from 500 to 1000 would decrease the CVs in the relative differences by about 29% (e.g., from 0.10 to 0.07) if the CVs are root n consistent (i.e., the CV decreases at a rate of $1/n^{1/2}$ as the number of draws, n, increases). Since the CVs given by 500 draws are already deemed to be acceptably low (e.g., G. Stauffer, AFSC, NMFS, Seattle, Wash.), this would not represent a significant improvement in precision.

© 1997 NRC Canada

Health and movements of fish in response to sediment sluicing in the Wind River, Wyoming[1]

Lee C. Bergstedt and Eric P. Bergersen

Abstract: A study of the effects of a sediment sluicing operation on the fish population of the Wind River, Wyoming, was conducted in 1991 and 1992. Sluicing from a lowhead diversion dam resulted in dramatic increases in suspended solids. Maximum suspended solid concentrations directly below the dam were as high as 18 000 mg/L (up to 1200 times mean concentrations recorded during normal low-flow conditions). Values from a sediment stress index indicated that sediment concentrations and durations were sufficient to cause behavioral, sublethal, and lethal responses in salmonids. A necropsy-based assessment of fish health showed significant signs of stress in mountain whitefish (*Prosopium williamsoni*) downstream of the dam. Negative physiological effects attributable to high suspended solid levels were seen in both internal and external examinations of the fish. Salmonid movement was measured by recapturing tagged fish. Fish above the dam moved an average of 1.8 km; those below the dam moved an average of 4.3 km. The increased movement of tagged salmonids below the dam was attributed to the sluicing operation. Fish population and community indices reflected a decline in species with specialized feeding and reproductive needs in the degraded habitat below the diversion dam.

Résumé : Une étude des effets d'une opération de lavage à l'auge (*sluicing*) des sédiments sur la population de poissons de la rivière Wind, au Wyoming, a été réalisée en 1991 et 1992. Le lavage effectué depuis un barrage de dérivation de faible chute a provoqué une augmentation spectaculaire de la concentration des solides en suspension qui, immédiatement en aval du barrage, s'élevait à une valeur maximale de 18 000 mg/L (jusqu'à 1200 fois les concentrations moyennes enregistrées au cours des périodes d'étiage normales). Les valeurs d'un indice de stress induit par les sédiments ont indiqué que les concentrations et la persistance des sédiments étaient suffisantes pour causer des réactions comportementales, sublétales et létales chez les salmonidés. Une évaluation par nécropsie de la santé du poisson a révélé des signes de stress substantiels chez le ménomini de montagnes (*Prosopium williamsoni*) en aval du barrage. Les effets physiologiques négatifs attribuables à des concentrations élevées de solides en suspension ont été observés aussi bien à l'examen interne qu'à l'examen externe des poissons. Les déplacements des salmonidés ont été mesurés par recapture de poissons marqués. Les poissons vivant en amont du barrage ont franchi en moyenne 1,8 km, tandis que les poissons vivant en aval se sont déplacés sur une distance moyenne de 4,3 km. Les déplacements plus importants des salmonidés marqués en aval du barrage ont été attribués à l'opération de lavage. Les indices des populations et communautés de poissons ont révélé un déclin chez les espèces ayant une alimentation et une reproduction spécialisées dans l'habitat dégradé situé en aval du barrage de dérivation.
[Traduit par la Rédaction]

Introduction

In 1934, a low-head dam (Diversion Dam) was constructed across the Wind River on the Wind River Indian Reservation, Wyoming, to divert water into the Midvale Irrigation District's Wyoming Canal. With construction of the dam and associated channel modifications, sediment was deposited in the river reach immediately upstream of Diversion Dam. These sediments impede the flow of water into the Wyoming Canal, and are routinely sluiced downstream by opening the dam gates (25 times in 1988 and 32 times in 1989; L.C. Bergstedt, unpublished data). Suspended solid levels during these events increased from as low as 2 mg/L to over 14 000 mg/L. Elevated suspended sediment levels can last up to 5 h directly below the diversion dam and 18 h 29 km downstream (L.C. Bergstedt, unpublished data). Similar concentrations and durations of suspended solids have been shown to cause negative effects in fish (Newcombe and MacDonald 1991). In addition, sediments are deposited downstream, further reducing the river's fishery potential by degrading spawning gravels and rearing habitat.

Previous sampling has shown that upstream of Diversion Dam the Wind River supports self-sustaining populations of rainbow trout (*Oncorhynchus mykiss*), brown trout (*Salmo trutta*), mountain whitefish (*Prosopium williamsoni*), and a mix of native fish including burbot (*Lota lota*), cyprinids, and catostomids (R.J. Baldes, unpublished data). Directly downstream of Diversion Dam, the fish population consists of native and introduced species including trout, burbot, and various catostomids and cyprinids.

Received January 25, 1995. Accepted July 31, 1996.
J12730

L.C. Bergstedt[2] and E.P. Bergersen. Colorado Cooperative Fish and Wildlife Research Unit, 201 Wagar, Colorado State University, Fort Collins, CO 80521, U.S.A.

[1] From a thesis submitted to the Academic Faculty of Colorado State University in partial fulfillment of the requirements of the degree of Master of Science.

[2] Current address: Montana Cooperative Fishery Research Unit, Montana State University, Bozeman, MT 59717-0346, U.S.A.

© 1997 NRC Canada

Methods

Study area

A 96.9-km reach of the Wind River in west central Wyoming was divided into two study sections. Locations reported here refer to river kilometres (rkm) from the Wedding of the Waters where the Wind River becomes the Bighorn River. The upstream section was from the western boundary of the Wind River Reservation (rkm 208.8) to Diversion Dam (rkm 147.5). The downstream section was from Diversion Dam to rkm 111.9.

General study design

The purpose of this investigation was to determine if sluicing activities were affecting the health and movement patterns of the fish in the Wind River. The general study design was to compare various measures of fish health above and below the diversion dam. Fish were collected using a raft-mounted electrofishing unit during three sampling runs from July through September in 1991 and 1992. All fish were identified to species and enumerated. All salmonids were weighed (±1 g) and measured (total length ± 1 mm), and locations were recorded to within 0.1 rkm. Salmonids greater than 200 mm were tagged with numbered Floy dart tags.

Suspended solids

Between June and September 1992, suspended solids were monitored upstream and downstream of the dam during normal low-water conditions, runoff periods, and sluicing events. Water samples were taken at various locations to determine the suspended solid concentration during these various events. Event durations were monitored while sampling. Water samples were kept cool until analyzed in the laboratory. A well-mixed sample of known volume was filtered, dried, and weighed using Standard Method 2540D (APHA 1989). A stress index was calculated by taking the natural logarithm of the product of concentration (milligrams per litre) and duration (hours) to predict the effects of the sluicing events on fish (Newcombe and MacDonald 1991).

Fish movements

Movements of salmonids greater than 200 mm total length were monitored by recording original capture locations and subsequent recapture locations to the nearest 0.1 km. Differences in upstream versus downstream movement between the two sections were tested with a Sign test. Differences between absolute distances moved above and below Diversion Dam were tested with a separate-variance *t*-test.

Organismic, population, and community indicators

Two organismic indicators of stress were used to evaluate the effects of sluicing on the fishery of the Wind River: the Fish Health/Condition Assessment Procedure (FHCAP, Goede and Barton 1990) and Fulton's condition factor (ktl, Cone 1989). Mountain whitefish were selected as the study species for the FHCAP because they are native, abundant, and representative of salmonids in how they express the effects of stressors in their environment (R. Goede, Fisheries Experiment Station, 1465 West 200 North, Logan, UT 84321, personal communication).

For the FHCAP, blood was sampled by cardiac puncture; differences in population means for hematocrit, leucocrit, and plasma protein were examined with a Student's *t*-test. External examination included eyes, gills, pseudobranchs, thymus, fins, and opercles. The examination listed the structures as normal or having some deviation from normal. An overall index value (Health Assessment Index) was then calculated based on a scoring system described by Adams et al. (1993). Differences between populations were tested with a chi-square test or a Mann–Whitney *U*-test if an index value was calculated.

The FHCAP internal examination included mesenteric fat, spleen, hindgut, kidney, liver, and bile, and comparisons between populations were made using the same methods as the external examinations.

Fulton's condition factor was used to evaluate fish condition for all salmonids and was expressed as ktl = [weight (g)/length $(mm)^3$] × 10^5 (Cone 1989). An analysis of variance (ANOVA) was used to test differences among sections.

Relative population density was used as an indicator of effects on the population. Catch per unit effort (CPUE) was used as a measure of density in the upstream and downstream sections. CPUE was calculated for mountain whitefish, brown trout, and rainbow trout in the upstream and downstream sections. ANOVA was used to test differences among sections.

Fish community feeding and reproductive guilds and tolerance to environmental degradation (Table 1) were also used to evaluate the effects of Diversion Dam. Guilds were defined from Schrader (1989) and Bramblett and Fausch (1991). Species not previously categorized were placed in guilds based on life history information found in Baxter and Stone (1995), Brown (1971), and Pflieger (1975). Feeding guilds were subdivided into omnivores and invertivores. The invertivore category included fish that include invertebrates for most of their diet, although the adults of some species may also consume fish for part of their diet. Reproductive guilds were categorized as generalist spawners and simple-lithophilous spawners (Balon 1975) or any other species believed to require gravel for spawning. Flathead chub (*Platygobio gracilis*) and mountain sucker (*Catostomus platyrhynchus*) were excluded from the analysis because of unknown reproductive behavior.

The common carp (*Cyprinus carpio*) and white sucker (*Catostomus commersoni*) were used as indicators of environmental degradation (Miller et al. 1988). Data analyses were performed using logistic regression. Significance tests were based on an *F*-statistic calculated from the chi-square values obtained for the between variance divided by within variance to provide a random effects test of section equivalent to a random effects test of ANOVA (Neter and Wasserman 1974).

Results

Suspended solids

Sluicing always produced dramatic increases in suspended solids. Mean suspended solid concentrations for normal low-flow conditions were 7.6 mg/L (SE = 2.1, n = 13) throughout the study area and 512.0 mg/L (SE = 46.5, n = 52) during rain and runoff conditions (L.C. Bergstedt, unpublished data). Postsluicing concentrations averaged 4034 mg/L (SE = 729.7, n = 36) directly below Diversion Dam and remained near 2000 mg/L 29 km downstream. The duration of the elevated levels of suspended solids lasted around 5 h directly below Diversion Dam and 18 h 29 km downstream. Suspended solid concentrations reached 1438.0 mg/L following a heavy rainstorm compared with a maximum concentration of nearly 18 000 mg/L directly below Diversion Dam during a sluicing event. Mean and maximum suspended solid concentrations below Diversion Dam during sluicing (Fig. 1) declined as the sediments moved downstream and were deposited in the river channel. These typical moving and declining maxima from Diversion Dam to the end of the downstream section are evident in the sluicing event that occurred on June 24, 1992 (Fig. 2). Stress Index values for all four locations during this sluice were near 10 (Table 2).

Fish movements

In 1991, 1035 fish were tagged in the study sections with 24 brown trout, 24 mountain whitefish, and 18 rainbow trout

© 1997 NRC Canada

Table 1. Feeding guild (I, invertivore; O, omnivore), reproductive guild (G, fishes requiring gravel for spawning; A, fishes with general spawning requirements; C, cavity nesters; U, unknown), and tolerance (T, tolerant) for fish species collected in the Wind River in 1991 and 1992 (from Baxter and Stone 1995; Brown 1971; Pflieger 1975; Schrader 1989; Bramblett and Fausch 1991).

Taxon	Feeding	Spawning	Tolerance
Family Salmonidae			
Oncorhynchus clarki bouvieri (Yellowstone cutthroat)[a]	I	G	
Oncorhynchus clarki (Snake River cutthroat)	I	G	
Oncorhynchus mykiss (rainbow trout)	I	G	
Prosopium williamsoni (mountain whitefish)[a]	I	G	
Salmo trutta (brown trout)	I	G	
O. mykiss × *O. clarki* hybrid (rainbow × cutthroat)	I	G	
Family Cyprinidae			
Couesius plumbeus (lake chub)[a]	I	A	
Cyprinus carpio (common carp)	O	A	T
Platygobio gracilis (flathead chub)[a]	I	U	
Rhinichthys cataractae (longnose dace)[a]	I	G	
Family Catostomidae			
Catostomus catostomus (longnose sucker)[a]	O	G	
Catostomus commersoni (white sucker)[a]	O	A	T
Catostomus platyrhynchus (mountain sucker)[a]	O	U	
Moxostoma macrolepidotum (shorthead redhorse)[a]	I	A	
Family Ictaluridae			
Noturus flavus (stonecat)[a]	I	C	
Family Gadidae			
Lota lota (burbot (ling))[a]	I	A	
Family Cottidae			
Cottus bairdi (mottled sculpin)	I	C	

[a] Denotes native species (Baxter and Stone 1995).

being recaptured. In 1992, 549 fish were tagged in the study sections with 19 brown trout, 23 mountain whitefish, and 9 rainbow trout being recaptured.

Recaptured salmonids in the upstream section moved less than those in the downstream section (mean = 1.8 km, SE = 0.33, $n = 47$ compared with mean = 4.3 km, SE = 0.75, $n = 70$). There was no indication of directional preference (upstream versus downstream movement; $P = 0.55$ by Sign test), but absolute distance moved was significant ($P = 0.001$ by separate-variance t-test (Fig. 3).

© 1997 NRC Canada

Fig. 1. Mean (SE) and maximum concentrations of suspended solids during sluicing events on the Wind River, Wyoming, in 1992 moving downstream from Diversion Dam.

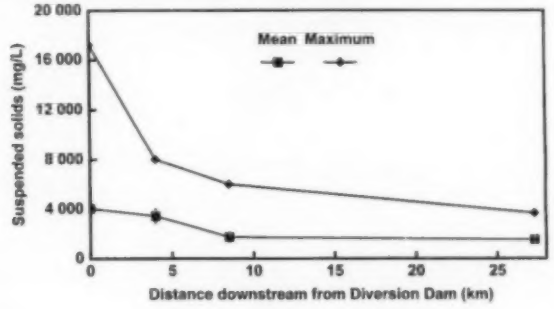
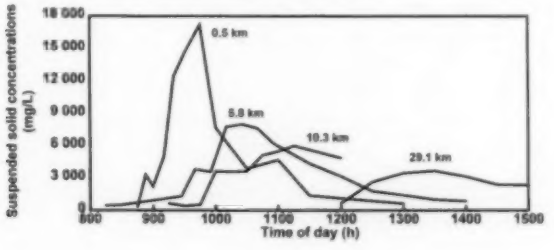


Fig. 2. Suspended solid concentrations over time at 0.5, 5.8, 10.3, and 29.1 km below Diversion Dam during the June 24, 1992, sluicing event on the Wind River, Wyoming.



Organismic, population, and community indicators

We performed necropsies on 21 adult mountain whitefish upstream of Diversion Dam between rkm 173.8 and 167.8 and 22 adult mountain whitefish downstream of Diversion Dam between rkm 147.4 and 141.8. The FHCAP (Table 3) showed that the mean hematocrit (% PCV) upstream ($n = 21$) was significantly higher than downstream ($n = 22$, $P = 0.014$ by Student's t-test). Mean leucocrit (% PCV) upstream was lower than downstream but not significantly. Plasma protein was significantly higher upstream than downstream ($P = 0.05$). External examination showed significantly higher percentages of abnormal gills, pseudobranchs, and fins downstream of Diversion Dam compared with upstream of Diversion Dam ($P = 0.013$, 0.0005, and 0.006, respectively, by chi-square test).

Internal examination showed that the mean mesenteric fat index upstream was significantly higher than downstream ($P = 0.0001$ by Mann–Whitney U-test). The spleen, hindgut, kidney, liver, and bile examinations showed no significant differences. The overall Health Assessment Index value upstream of Diversion Dam was significantly higher than downstream ($P = 0.01$ by Mann–Whitney U-test).

Mean condition (ktl) of mountain whitefish, brown trout, and rainbow trout was significantly higher ($P = 0.0001$, 0.0001, and 0.02, respectively, by ANOVA) upstream than downstream in 1991. In 1992, mean ktl values for all three species were again higher upstream but the difference for rainbow trout was not significant ($P = 0.48$) (Table 4).

In 1991 the mean CPUE for brown trout and rainbow trout

Fig. 3. Capture and recapture locations of marked brown trout, rainbow trout, and mountain whitefish in the Wind River, Wyoming, 1991 and 1992. The diagonal line indicates zero net movement.

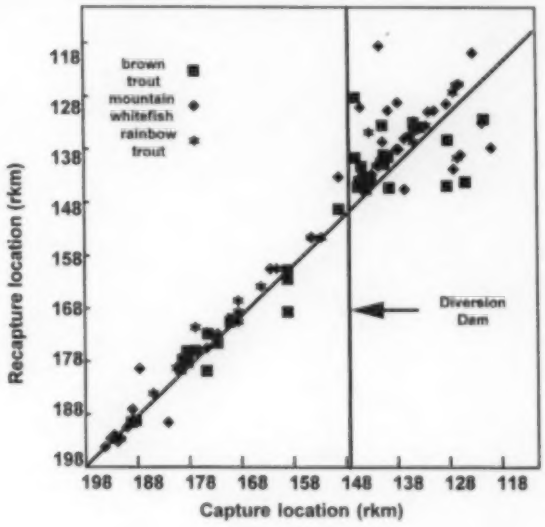
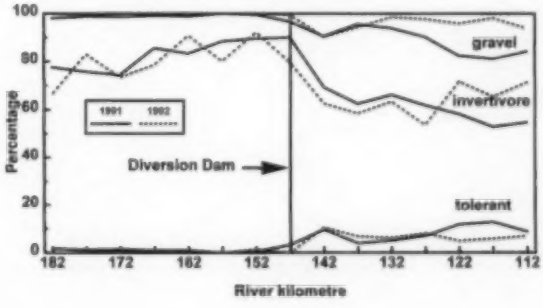


Fig. 4. Percentage of fish in total catch belonging to invertivore feeding guild and gravel spawning reproductive guild, and fish tolerant to habitat degradation above and below Diversion Dam on the Wind River, Wyoming, in 1991 and 1992.



was significantly higher upstream than downstream ($P = 0.01$, $P = 0.04$ by ANOVA), but mountain whitefish CPUE was not ($P = 0.35$). In 1992, CPUE was not significantly different between upstream and downstream sections for any of the three species (Table 5; $P = 0.74$ for mountain whitefish, $P = 0.98$ for brown trout, and $P = 0.39$ for rainbow trout). The fall sampling run was excluded from the analysis because the Diversion Dam acted as a fish barrier and concentrated brown trout and mountain whitefish directly below it.

In 1991 the proportions of invertivores and species requiring gravel for spawning (Fig. 4) upstream were significantly higher than downstream ($P = 0.0001$, logistic regression). The proportion of species tolerant to habitat degradation was significantly lower upstream than downstream of Diversion Dam ($P = 0.0001$). Similar trends were observed in 1992 (Fig. 4), but the proportion of invertivores was not significantly higher upstream than downstream ($P > 0.25$).

© 1997 NRC Canada

Table 2. Mean suspended solid concentrations (mg/L), durations (h), and stress index values (Newcombe and MacDonald 1991) 0.5, 5.8, 10.3, and 29.1 km below Diversion Dam on the Wind River, Wyoming, during the June 24, 1992, sluicing event.

Location	Mean concentration	Duration	Stress index value
0.5	4183.6	4.8	9.9
5.8	2922.1	6.0	9.8
10.3	2650.0	9.3	10.1
29.1	1698.1	18.7	10.4

Table 3. Mean (SE) measurements, percentages of normal fish, or average index values for blood constituents and body and organ conditions of mountain whitefish from above and below Diversion Dam.

Feature measured	Upstream	Downstream
Hematocrit (% PCV)	53.53(1.39)	48.67(1.59)*
Coefficient of variation	11	15
Leucocrit (% PCV)	0.90(0.10)	1.05(0.12)
Plasma protein	5.12(0.20)	4.65(0.19)*
Eyes (% normal)	100	100
Gills (% normal)	86	55*
Pseudobranchs (% normal)	76	27*
Thymus (% normal)	29	50
Thymus index	0.71	0.54
Fins (% normal)	95	63*
Fin damage index	0.05	0.36*
Opercles (% normal)	100	100
Opercles shortening index	0.00	0.00
Mesenteric fat index	3.00	1.77*
Spleen (% normal)	90	95
Hind gut (% normal)	67	55
Hind gut index	0.33	0.45
Kidney (% normal)	100	100
Liver (% normal)	71	77
Bile color index	1.40	1.59
Mean Health Assessment Index	65.5	42.9
Sample size	21	22

Note: Asterisks indicate significant differences ($P < 0.05$) from upstream values. Means compared by Student's *t*-test, percent-normal values using chi-square tests, and average index values compared by Mann–Whitney *U*-tests. PCV is packed cell volume.

Discussion

Suspended solids

During sluicing events, maximum suspended solid concentrations directly below Diversion Dam were up to 1200 times the mean suspended solid concentrations seen during clear water conditions. Suspended solids can have a variety of direct and indirect effects on a river and its biota. Direct effects on freshwater fish include mortality, reduced growth rates, or reduced resistance to disease (Alabaster and Lloyd 1982). Other possible effects include abnormal development of eggs and larvae, alteration of movements and migrations, and a reduction in the abundance of forage organisms. Suspended solids have also been shown to adversely affect aquatic invertebrates (reviewed by Newcombe and MacDonald 1991) and periphyton (Alabaster and Lloyd 1982).

Although the effects of suspended solids on fish have been well studied, there is a lack of agreement on the concentrations necessary to cause these effects. Everest et al. (1987) reported that mortality of salmonids only occurs when concentrations are greater than 20 000 mg/L. Newcombe and MacDonald (1991) suggested that mortality can occur at lower concentrations and that the severity of the effect is also related to the duration of the event. From an extensive literature review, they developed a stress index that takes both concentration and duration into account. Others have suggested that threshold levels, temperature, fish size, and particle size are also important variables (Servizi and Martens 1992; Gregory et al. 1993). Although there is some uncertainty in what suspended solid levels cause various responses, stress index values calculated for the Wind River during sluicing events have been reported to cause behavioral, sublethal, and lethal responses in salmonids (Newcombe and MacDonald 1991).

Fish movements

Recaptured fish moved more below Diversion Dam than above. Sluicing activities could cause an increase in movement by eliciting an avoidance response or by obscuring the visual cues by which fish maintain position (Berg and Northcote 1985). Doeg and Koehn (1994) reported dramatic decreases in blackfish populations directly downstream from a weir that was being desilted at suspended sediment concentrations of 4610 mg/L. Berg and Northcote (1985) reported territorial and feeding behavior disruptions in juvenile coho salmon (*Oncorhynchus kisutch*) as a result of avoidance responses to short-term pulses of suspended sediments in an experimental stream. They suggested that these behavioral responses impaired the fitness of juvenile fish frequently subjected to suspended sediment pulses. These behavioral responses may decrease fitness in the adult salmonid population on the Wind River below Diversion Dam as well.

Organismic, population, and community indicators

The FHCAP provided evidence that fish below Diversion Dam were adversely affected by sluicing events. Blood constituent measures can be useful in assessing negative effects (Goede and Barton 1990), and hematocrit values recorded for mountain whitefish on the Wind River were at the upper end of values observed for other salmonids in other studies (Cardwell and Smith 1971; McLeay and Gordon 1977; Wells et al. 1984; Barton et al. 1985; Goede and Barton 1990). High hematocrit may be the result of acute stress (Barton et al.

© 1997 NRC Canada

Table 4. Fulton's condition index comparisons for mountain whitefish, brown trout, and rainbow trout between upstream and downstream sections on the Wind River in 1991 and 1992.

	1991		1992	
Fish species	Upstream	Downstream	Upstream	Downstream
Mountain whitefish	1.04	1.01*	0.98	0.93*
Brown trout	1.16	1.03*	1.12	1.06*
Rainbow trout	1.10	1.06*	1.05	1.03

Note: Asterisks denote significant differences between sections for that year ($P < 0.05$ by ANOVA).

Table 5. Catch per unit effort (adult fish per hour) for mountain whitefish, brown trout, and rainbow trout upstream and downstream of Diversion Dam from July through October in 1991 and 1992.

	1991		1992	
Fish species	Upstream	Downstream	Upstream	Downstream
Mountain whitefish	9.6	7.2	13.8	16.2
Brown trout	5.4	2.4*	7.8	7.8
Rainbow trout	2.4	1.2*	4.8	3.0

Note: Asterisks denote significant differences between sections for that year ($P < 0.05$ by ANOVA).

1985), but ranges of hematocrit values for unstressed mountain whitefish have not been reported. The coefficient of variation (15) for hematocrit (% PCV) in the downstream section was high enough to suggest that the population may be unhealthy (Goede and Barton 1990). Leucocrit and plasma protein values were within the normal range reported for other salmonids (McLeay and Gordon 1977; Wedemeyer et al. 1983; Cunjak 1988; Goede and Barton 1990).

The examination of external features showed that the fish downstream of Diversion Dam were being negatively effected by sluicing. Significantly more fish had abnormal gills below than above the dam. Suspended solids can interrupt gill ventilation and cause "coughing." Coughing has been used as an indicator of sublethal stress in some salmonids (Berg and Northcote 1985; Servizi and Martens 1992). Fine silt and clay can clog gills (Gordon et al. 1992) and interrupt gas exchange.

Only 27% of the fish examined downstream of the dam appeared to have normal pseudobranchs compared with 76% above the dam. Although Goede and Barton (1990) interpreted the swelling of the pseudobranch as an indication of changes in oxygen and carbon dioxide partial pressure, in the case of the Wind River, it may also be the result of abrasion by the high suspended solid concentrations.

Although only mild fin erosion was observed, fin damage was significantly higher downstream than upstream of the dam. Physical factors such as abrasion were the most probable cause, although chemical factors and disease may also cause erosion (Goede and Barton 1990).

The mean mesenteric fat index was the only internal component of the necropsy found to be significantly lower downstream of the dam. Mesenteric fat deposits are not directly related to stress but indicate feeding intensity and energy deposition (Goede and Barton 1990).

The necropsy provided evidence that the mountain whitefish below Diversion Dam were experiencing more stress than those above the dam. Since flows measured in these two sections (L.C. Bergstedt, unpublished data) were well above the minimum flow recommendations of the Cooperative Instream Flow Service Group (Vogel 1980), and temperature and conductivity measurements did not differ significantly between sections (L.C. Bergstedt, unpublished data), it appears that the elevated suspended solids produced during sluicing are adversely affecting salmonids.

Differences in condition factors also indicate that fish downstream of the dam were not as healthy as those upstream. Declines in condition indicate lower energy reserves (Goede and Barton 1990). This can be the result of altered feeding patterns (Brown et al. 1987) or increased metabolic rates resulting from stress (Barton and Schreck 1987). In both years, mean ktl values were higher above than below the dam. Only rainbow trout condition in 1992 was not significantly lower below the dam.

CPUE was significantly higher above than below the dam in 1991 for brown trout and rainbow trout when late fall sampling was excluded. Various factors may act together to depress fish populations below the dam. Direct mortality at all life stages, reductions in suitable spawning habitat, and a reduction in forage organisms may occur concurrently.

Guilds have been used as indicators of many types of environmental degradation across spatial and temporal

© 1997 NRC Canada

scales (Fausch et al. 1990). The main advantages of using guilds as indicators of environmental degradation are that they are simple and allow identification of specific stresses. We chose to look at feeding and reproductive guilds because they are both sensitive to siltation (Berkman and Rabeni 1987). The percentages of invertivores and fish requiring gravel for spawning decreased below Diversion Dam. The increase in the percentage of omnivores and the concurrent decrease in the percentage of invertivores may be the result of decreased invertebrate production resulting from increased sedimentation below Diversion Dam (Doeg and Koehn 1994). Increased sedimentation would also explain the decrease in the percentage of fish requiring gravel for spawning below the dam. The proportion of tolerant species also increased below Diversion Dam.

Natural history studies are often the only tool available in ecological impact assessments, and care must be taken in planning the study and interpreting the results. Use of the before-after-control-impact model (Stewart-Oaten et al. 1986; Smith et al. 1993) has alleviated some of these concerns, but in the case of the Wind River, no data exist before the construction of Diversion Dam. Because of these study limitations, caution must be used in the inferences made of any statistical analysis used.

This study provided evidence that the sediment sluicing operations were adversely affecting the fish community on the Wind River. By conducting a study that examined various levels of biological organization simultaneously, we were able to determine the magnitude of the effects. Necropsy and fish condition data suggest detrimental effects to the individual organism, and population and community structure data show that species with specialized feeding and reproductive needs decline below Diversion Dam.

Acknowledgments

We thank Kurt D. Fausch and John D. Stednick for their input and comments throughout the course of this study. We thank Richard J. Baldes, David L. Skates, Dan Stubbs, Jeff Kimber, Laurie Connell, Misti Knodell, and Crystal Hudson for help in sampling. We thank Gary White and Bob McFarland for their assistance in data analysis. We also thank Robert G. White, Charles Gowan, and two anonymous reviewers for review of the manuscript. This research was funded and supported by the Shoshone and Arapaho tribes of the Wind River Indian Reservation and the U.S. Environmental Protection Agency. Cooperators in the Colorado Cooperative Fish and Wildlife Research Unit include the National Biological Service, Colorado Division of Wildlife, and Colorado State University.

References

Adams, S.M., Brown, A.M., and Goede, R.W. 1993. A quantitative health assessment index for rapid evaluation of fish condition in the field. Trans. Am. Fish. Soc. **122**: 63–73.

Alabaster, J.S., and Lloyd, R. 1982. Finely divided solids. *In* Water quality criteria for freshwater fish. 2nd ed. *Edited by* J.S. Alabaster and R. Lloyd. Butterworth, London. pp. 1–20.

APHA (American Public Health Association, American Water Works Association, and Water Pollution Control Federation). 1989. Standard methods for the examination of water and wastewater. 17th ed. APHA, Washington, D.C.

Balon, E.K. 1975. Reproductive guilds of fishes: a proposal and definition. J. Fish. Res. Board Can. **32**: 821–864.

Barton, B.A., and Schreck, C.B. 1987. Metabolic cost of acute physical stress in juvenile steelhead. Trans. Am. Fish. Soc. **116**: 257–263.

Barton, B.A., Weiner, G.S., and Schreck, C.B. 1985. Effect of prior acid exposure on physiological responses of juvenile rainbow trout (*Salmo gairdneri*) to acute handling stress. Can. J. Fish. Aquat. Sci. **42**: 710–717.

Baxter, G.T., and Stone, M.D. 1995. Fishes of Wyoming. Wyoming Game and Fish Department, Cheyenne, Wyo.

Berg, L., and Northcote, T.G. 1985. Changes in territorial, gill-flairing, and feeding behavior in juvenile coho salmon (*Oncorhynchus kisutch*) following short-term pulses of suspended sediment. Can. J. Fish. Aquat. Sci. **42**: 1410–1417.

Berkman, H.E., and Rabeni, C.F. 1987. Effect of siltation on stream fish communities. Environ. Biol. Fishes, **18**: 285–294.

Bramblett, R.G., and Fausch, K.D. 1991. Variable fish communities and the index of biotic integrity in a western Great Plains river. Trans. Am. Fish. Soc. **120**: 752–769.

Brown, C.J.D. 1971. Fishes of Montana. Big Sky Books, Montana State University, Bozeman, Mont.

Brown, J.A., Johansen, P.H., Colgan, P.W., and Mathers, R.A. 1987. Impairment of early feeding behavior of largemouth bass by pentachlorophenol exposure: a preliminary assessment. Trans. Am. Fish. Soc. **116**: 71–78.

Cardwell, R.D., and Smith, L.S. 1971. Hematological manifestations of vibriosis upon juvenile chinook salmon. Prog. Fish-Cult. **33**: 232–235.

Cone, R.S. 1989. The need to reconsider the use of condition indices in fishery science. Trans. Am. Fish. Soc. **118**: 510–514.

Cunjak, R.A. 1988. Physiological consequences of overwintering in streams: the cost of acclimatization? Can. J. Fish. Aquat. Sci. **45**: 443–452.

Doeg, T.J., and Koehn, J.D. 1994. Effects of draining and desilting a small weir on downstream fish and macroinvertebrates. Regul. Rivers Res. Manage. **9**: 263–277.

Everest, F.H., Beschta, R.L., Scrivener, J.C., Doski, D.V., Sedell, J.R., and Cederholm, C.J. 1987. Fine sediments and salmonid production: a paradox. *In* Streamside management: forestry and fishery interactions. *Edited by* E.O. Salo and T.W. Cundy. Contract No. 57. Institute of Forest Research, University of Washington, Seattle, Wash. pp. 98–142.

Fausch, K.D., Lyons, J., Karr, J.R., and Angermeier, P.L. 1990. Fish communities as indicators of environmental degradation. Am. Fish. Soc. Symp. **8**: 123–144.

Goede, R.W., and Barton, B.A. 1990. Organismic indices and an autopsy-based assessment as indicators of health and condition of fish. Am. Fish. Soc. Symp. **8**: 93–108.

Gordon, N.D., McMahon, T.A., and Finlayson, B.L. 1992. Stream hydrology: an introduction for ecologists. John Wiley & Sons, Chichester, England.

Gregory, R.S., Servizi, J.A., and Martens, D.W. 1993. Comment: utility of the stress index for predicting suspended sediment effects. N. Am. J. Fish. Manage. **13**: 868–873.

McLeay, D.J., and Gordon, M.R. 1977. Leucocrit: a simple hematological technique for measuring acute stress in salmonid fish, including stressful concentrations of pulpmill effluent. J. Fish. Res. Board Can. **34**: 2164–2175.

Miller, D.L., Leonard, P.M., Hughes, R.M., Karr, J.R., Moyle, P.M., Schrader, L.H., Thompson, B.A., Daniels, R.A., Fausch, K.D., Fitzhugh, G.A., Gammon, J.R., Halliwell, D.B., Angermeier, P.L., and Orth, D.J. 1988. Regional applications of an index of biotic integrity for use in water resource management. Fisheries, **13**(5): 12–20.

Neter, J., and Wasserman, W. 1974. Applied linear statistical models. Richard D. Irwin, Inc., Homewood, Ill.

© 1997 NRC Canada

Newcombe, C.P., and MacDonald, D.D. 1991. Effects of suspended sediments on aquatic ecosystems. N. Am. J. Fish. Manage. **11**: 72–82.

Pflieger, W.L. 1975. The fishes of Missouri. Missouri Department of Conservation, Jefferson, Mo.

Schrader, L.H. 1989. Use of the index of biotic integrity to evaluate fish communities in western Great Plains streams. M.Sc. thesis, Colorado State University, Fort Collins, Co.

Servizi, J.A., and Martens, D.W. 1992. Sublethal responses of coho salmon (*Oncorhynchus kisutch*) to suspended sediments. Can. J. Fish. Aquat. Sci. **49**: 1389–1395.

Smith, E.P., Orvos, D.R., and Cairns, J. Jr., 1993. Impact assessment using the before-after-control-impact (BACI) model: concerns and comments. Can. J. Fish. Aquat. Sci. **50**: 627–637.

Stewart-Oaten, A., Murdoch, W.W., and Parker, K.R. 1986. Environmental impact assessment: pseudoreplication in time? Ecology, **67**: 929–940.

Vogel, D.A. 1980. Instream flow recommendations for the fishery resources in the major rivers and streams on the Wind River Indian Reservation, Wyoming. U. S. Fish and Wildlife Service, Lander, Wyo.

Wedemeyer, G.A., Gould, R.W., and Yasutake, W.T. 1983. Some potentials and limits of the leucocrit test as a fish health assessment method. J. Fish Biol. **23**: 711–716.

Wells, R.M.G., Tetens, V., and Devries, A.L. 1984. Recovery from stress following capture and anaesthesia of antarctic fish: haematology and blood chemistry. J. Fish Biol. **25**: 567–576.

© 1997 NRC Canada

Impacts of a herbicide and fertilizers on the microbial community of a saline prairie lake

Marley J. Waiser and Richard D. Robarts

Abstract: Prairie wetlands are scattered among cultivated land where herbicides and fertilizers are regularly used. Avadex-BW© (active ingredient triallate) is one of the most popular pre-emergent herbicides used on the Canadian prairies to control wild oats and other broadleaf weeds. Triallate enters aquatic systems through processes of runoff and direct overspray, while fertilizers (nitrogen and phosphorus) enter through runoff and aeolian deposition. Microcosm studies on Redberry Lake, an oligotrophic prairie lake, indicated that additions of triallate stimulated bacterial production ([^{3}H]thymidine incorporation), metabolism ([^{14}C]glucose uptake), and numbers, but only if nitrogen and phosphorus were added. Because the microbial community is nutrient limited, there is the possibility that triallate could bioaccumulate in this system. Addition of nitrogen and phosphorus (N + P) stimulated measures of phytoplankton biomass (Chl *a*), bacterial metabolism, numbers, and production. Although triallate had little effect on bacteria, at any concentration tested, substantial declines in phytoplankton biomass occurred at triallate concentrations $\geq$1000 $\mu g \cdot L^{-1}$. Greatest impacts of herbicides and nutrients, however, are expected in spring as a result of runoff events, aeolian deposition, and direct overspray.

Résumé : Les terres humides des prairies sont dispersées parmi les terres cultivées dans lesquelles on utilise régulièrement des engrais et des herbicides. Avadex-BW© (dont l'ingrédient actif est le triallate) est l'un des herbicides de pré-levée les plus fréquemment employés dans les prairies canadiennes pour lutter contre les avoines sauvages et d'autres mauvaises herbes dicolylédones. Le triallate se répand dans les systèmes aquatiques par ruissellement et par pulvérisation hors cible directe, alors que les engrais (azote et phosphore) le font par ruissellement et dépôt éolien. Des études en microcosmes effectuées dans le lac Redberry, un lac oligotrophe des prairies, ont indiqué que l'addition de triallate stimulait la production (incorporation de [^{3}H]thymidine), le métabolisme (absorption de [^{14}C]glucose) et le nombre de bactéries uniquement lorsqu'on ajoute de l'azote et du phosphore. Comme la communauté microbienne dispose d'un apport limité en éléments nutritifs, il est possible que le triallate s'accumule dans ce système. L'addition d'azote et de phosphore (N + P) a fait augmenter les valeurs relatives à la biomasse de phytoplancton (Chl *a*), au métabolisme, à la production et au nombre de bactéries. Bien que le triallate n'avait que peu d'effet sur les bactéries, pour toutes les concentrations testées, une diminution substantielle de la biomasse de phytoplancton a été observée lorsque la concentration de triallate était supérieure ou égale à 1000 $\mu g \cdot L^{-1}$. Cependant, on prévoit que les répercussions les plus importantes des herbicides et éléments nutritifs surviennent au printemps par suite du ruissellement, du dépôt éolien et de la pulvérisation hors cible directe.
[Traduit par la Rédaction]

Introduction

The myriad of wetlands, including saline lakes, found within the prairie and Great Plains ecozone produce 50–75% of North American waterfowl in any given year (Mitsch and Gosselink 1993; Donald and Syrgiannis 1995). Wetlands across the prairies are regularly interspersed among cultivated fields where both fertilizers and pesticides are commonly used. About 10^6 kg of herbicides is applied annually to agricultural land across the prairies, with half of this usage occurring in Saskatchewan (Waite et al. 1995). Herbicides and fertilizers make their way into waterbodies through processes of runoff, groundwater flow, spray drift, or aeolian deposition (Grover et al. 1988; Waite et al. 1992). Moreover, on the prairies where a high level of soil fertility is maintained by fertilizing, nutrient enrichment of surface waters may be considerable, especially at certain times of the year. Although the potential effects of these chemicals (and nutrients) on prairie wetlands are largely unknown (Johnson 1986), it has been hypothesized that pesticides might adversely affect primary production, thus negatively affecting zooplankton and avian production (Donald and Syrgiannis 1995).

The herbicide most widely used on the Canadian prairies to control wild oats and other broadleaf weeds is AVADEX-BW©, a pre-emergent herbicide first introduced to the Canadian market in 1962. In 1989, 3295 $\times$ 10^3 kg of AVADEX-BW© was applied to cultivated fields in Saskatchewan (Lewis 1991). The active ingredient in this herbicide is triallate (S,2,3-trichloroallyl diisopropyl), comprising 40.38% of the formulation. The remaining inert ingredients include C-9 aromatics (53.5%), emulsifiers (4.5%), and others (1.62%) (Material Safety Data Sheet, Monsanto Canada Inc.). Triallate has a molecular weight of 304.7 and water solubility of 4 $mg \cdot L^{-1}$ (at 25°C), and with an octanol–water partition coefficient (log K_{ow}) of 4.6, it is moderately lipophilic (Humburg 1989). It also has a C:N ratio of ~9:1 and is phosphorus free.

Received January 9, 1996. Accepted July 15, 1996.
J13248

M.J. Waiser and R.D. Robarts. Environmental Sciences Division, National Hydrology Research Institute, Environment Canada, 11 Innovation Boulevard, Saskatoon, SK S7N 3H5, Canada.

© 1997 NRC Canada

AVADEX-BW© is marketed as an amber oil but is also available in granular formulation. Because of the extremely volatile nature of the oil-based application, this herbicide must be harrowed into the soil immediately after spraying (Environment Canada 1992). Despite harrowing, volatilization losses as high as 17.6% of the amount of triallate applied have been reported in Saskatchewan in a single growing season, with 50% of this loss occurring during the first 4–5 days after application (Grover et al. 1988). Volatilized or adsorbed to airborne soil particles, triallate eventually makes its way into surface waters. In the granular formulation, triallate is applied to fields in the fall and may be left unincorporated on the soil surface until spring. In the spring, most of the triallate loss from prairie agriculture watershed can be traced to spring runoff of triallate applied the previous fall (Williamson 1984). Concentrations of triallate ranging from 1.58 to 6.77 $\mu g \cdot L^{-1}$ have been detected in spring runoff and snowmelt in Saskatchewan (Waite et al. 1986).

Despite being one of the most widely used herbicides on the prairies today, there is a startling lack of research into the effects of triallate on aquatic food web components. Among the two published studies found, Johnson (1986) showed that triallate may be directly toxic to certain species of aquatic invertebrates, which in turn are a critical food source for reproducing ducks. Because of its negative effects on phytoplankton as well, Johnson (1986) suggested that caution be used in the application of triallate in or near wetland habitats. Arts et al. (1995) assessed the potential for uptake of triallate (a lipophilic contaminant) into lipid-rich tissues of two aquatic invertebrates (*Hyallella azteca* and *Gammarus lacustris*). According to this study, triallate was mainly stored in the triacylglycerol energy reserves of these animals. Moreover, triacylglycerol stores were highest in spring and fall, which coincided with the major application times for triallate. To date, there has been no research into the effects of this herbicide on the bacterial component of the pelagic aquatic food web.

Our study considers the effects of nutrients (nitrogen and phosphorus) and the herbicide Avadex BW© (triallate, Monsanto Corporation, St. Louis, Mo.) on the structure and function of the microbial community of Redberry Lake, an oligotrophic saline lake located in south-central Saskatchewan. In particular, it examines the effects of this herbicide on measures of phytoplankton biomass (Chl *a*), bacterial numbers, bacterial metabolism ([^{14}C]glucose uptake), and bacterial production ([^{3}H]thymidine incorporation). We expected that nutrients would have a positive effect on bacteria and phytoplankton and that the herbicide would have a negative effect on phytoplankton and no effect on bacteria. We were also interested in the combined effects of nutrients and herbicides: could the combination overcome a negative effect of herbicide on phytoplankton and was there a possibility of a synergistic effect for bacteria? To investigate these effects, a microcosm approach, using natural microbial assemblages, was used. Such microcosms are useful for this purpose because in contrast with single-species bioassays, they display community and ecosystem level processes, even though the magnitude and rate of these processes may not exactly mirror the naturally occurring ecosystem (Giesy and Allred 1985). In addition, microcosms are easily replicated and are practical in terms of costs and logistics (Van Donk et al. 1995).

Materials and methods

Study site

Redberry Lake (Z_{max} = 16 m, surface area 45 km^2) is an oligotrophic (mean summer Chl *a* of 0.7 $\mu g \cdot L^{-1}$) lake situated on a glacial lacustrine alluvial plain in south-central Saskatchewan (52°43′N, 107°09′W). Except for a narrow buffer zone of brush, aspen forest, and grassland, the lake is totally surrounded by cultivated land. Redberry Lake is saline (total dissolved solids (TDS) 20.9 $g \cdot L^{-1}$), with magnesium (68.1%) and sodium (29.2%) cations and sulfate (93.7%) anions dominating (Arts et al. 1993). Although levels of total and soluble reactive phosphorus in the lake are high (46.5 and 16 $\mu g \cdot L^{-1}$, respectively), little is available for microbial uptake, and consequently, the microbial population shows strong signs of phosphorus limitation (Waiser and Robarts 1995). Concentrations of NO_2+NO_3-N are usually <2.0 $\mu g \cdot L^{-1}$, and NH3-N concentrations are mostly <30 $\mu g \cdot L^{-1}$ during the ice-free season (Robarts et al. 1992). Despite dissolved organic carbon (DOC) concentrations of 37 $mg \cdot L^{-1}$, the lake water remains clear. Mean Z_{eu} (euphotic zone depth) for this lake is 15.2 m (Robarts et al. 1992).

Generally, the significance of microbial processes increases along a gradient leading from eutrophy to oligotrophy (Cole et al. 1988). In an oligotrophic system such as Redberry Lake, picoplankton and heterotrophic bacteria may be key components in carbon metabolism and energy transfer (cf. Simon et al. 1992). Any perturbation, such as nutrients or herbicides that might alter microbial processes, has the potential to alter carbon and energy flow in the ecosystem as well. Because Redberry Lake is located in an endorheic (hydrologically closed) drainage basin, any herbicides or nutrients entering this system as a result of agricultural practices will stay in the basin. It is therefore important to quantify the possible effects that the addition of herbicides and nutrients might have on the microbial community of prairie saline lakes such as Redberry.

Sample collection

Water for these experiments was collected from the epilimnion of Redberry Lake and placed in three carboys. Carboys were cleaned with ethanol, rinsed with Milli-Q water, and then thoroughly rinsed with lake water before filling. The filled carboys were kept in the dark, on ice, during transport back to the laboratory.

At the laboratory, the water was screened through 153-μm pore-size mesh (Nitex) to remove large zooplankters. To establish initial levels of Chl *a*, bacterial numbers, bacterial production, and metabolism, aliquots of water from each carboy were removed and processed in the following fashion.

One litre of water was filtered through GF/C filters and frozen for subsequent fluorometric Chl *a* analysis using the 90% boiling ethanol technique (Nusch 1980). From each carboy, one 10-mL sample was removed, placed in a sterile 20-mL Vacutainer tube, and preserved with 200 μL of Lugol's solution (Pomroy 1984) for subsequent bacterial counts using DAPI and epifluorescent microscopy (Porter and Feig 1980). Lugol's and DAPI solutions were prefiltered through 0.2-μm pore-size cellulose nitrate filters before use.

In this study, the rate of uptake and respiration (v) of uniformly labelled high specific activity glucose (>8.5 $GBq \cdot mmol^{-1}$) at a concentration of 50 $\mu g\ C \cdot L^{-1}$ was

© 1997 NRC Canada

used as a measure of relative heterotrophic activity (Hobbie and Crawford 1969; Wright 1978). Incubation time was 1 h. Uptake over this time was linear and the glucose concentration used saturated bacterial uptake. From each carboy, two 10-mL samples (one live and one control) were aseptically withdrawn, processed, and filtered according to the method of Hobbie and Crawford (1969). Samples were then counted in a Packard 1900 CA liquid scintillation spectrometer with automatic quench correction. Rates of [^{14}C]glucose uptake (v) were calculated according to Wright (1978). Mean and standard error were then computed for each of the treatments. In addition, rates of glucose uptake were divided by bacterial numbers to give micrograms of glucose ($\times 10^{-11}$) taken up per cell per hour.

Samples were also withdrawn and analysed for measures of bacterial production as indicated by the rate of [methyl-^{3}H]thymidine (TdR) incorporation into bacterial DNA. Two 10-mL samples (one live and one control) were aseptically withdrawn from each carboy and placed into 15-mL sterile screw-top glass tubes. Controls were prepared by addition of 0.5 mL of formaldehyde, followed by 0.5 mL of 5 M NaOH (M.J. Waiser, unpublished data). TdR was added to each sample to a final concentration of 20 nM. Samples were incubated in an environmental chamber in the dark and at lake temperature for 1 h. Prior experiments (Robarts and Zohary 1993) indicated that 20 nM TdR was saturating and that uptake at this concentration was linear over a 2-h period. At the end of 1 h, live samples were killed in the same manner as controls and then labelled DNA was extracted according to Robarts and Wicks (1989). All samples were counted in a Packard 1900 CA scintillation spectrometer with automatic quench correction. TdR incorporation was calculated for each carboy (live – control) and the mean and standard error were calculated. In addition, TdR incorporation was divided by bacterial numbers and expressed as picomoles ($\times 10^{-9}$) of TdR incorporated per cell per hour.

After establishing initial Chl *a* concentrations, bacterial numbers, and rates of incorporation of [^{3}H]thymidine and uptake of [^{14}C]glucose for each carboy, microcosm experiments were initiated.

Microcosm experiments

Two microcosm experiments were run: one in August 1992 and the other in September 1992.

Microcosm experiment, August 1992

Each microcosm consisted of 1000 mL of prescreened lake water, containing the natural microbial assemblage, in a 1000-mL autoclaved Erlenmeyer wide-mouth flask. All glassware used in the experiment was acid washed, thoroughly rinsed with Milli-Q water, and dried before autoclaving.

Eight treatments were chosen and each of the treatments was run in triplicate. Each water sample of the triplicate, moreover, was taken from a different carboy to avoid pseudoreplication (Hurlbert 1984). Nitrogen and phosphorus stock solutions (as $NaNO_3$ and KH_2PO_4) were prepared in autoclaved Milli-Q water and added to the microcosms to achieve a mole ratio of 36:1 (Elser et al.1990). A triallate stock solution of 4.8 g·L^{-1} was prepared by diluting 0.9 mL of Avadex (400 g·L^{-1} as triallate) in 75 mL of acetone. The treatments were as follows: control (no addition), + nitrogen + phosphorus (N + P), three concentrations of triallate (10, 100, and 1000 μg·L^{-1}), and the same three concentrations of herbicide but with nitrogen and phosphorus added to each. The dosage was reflective of possible herbicide runoff values, which could vary from nil to 1000 μg·L^{-1} or more (Johnson 1986). Resulting percentages (v/v) of acetone added to the microcosms were 0.02% (1000 μg·L^{-1} treatment), 0.002% (100 μg·L^{-1} treatment), and 0.0002% (10 μg·L^{-1} treatment). These acetone concentrations were deemed well below the level that would have an adverse effect on the microbial population (Brown and Lean 1995; Arts et al. 1995).

Nutrients and herbicides were pipetted into each of the microcosms and the water was well mixed with a glass stirring rod. Each of the microcosms was closed with an autoclaved styrofoam plug and all vessels were transferred to a large shaking table located in an environmental chamber. The samples were incubated at lake temperature in the dark on a 12 h light : 12 h dark cycle for 4 days. Light levels in the incubator were 90 μE·m^{-2}·s^{-1} as measured with a quantum sensor and meter (LiCor, model 1900B). At about 24-h intervals, samples were aseptically withdrawn from each of the microcosms, preserved for bacterial numbers, and processed for TdR incorporation and [^{14}C]glucose uptake as noted above. For each flask, one live and one control sample was run for TdR and [^{14}C]glucose uptake. The August experiment was run for 4 d.

At the end of the experiment, the volume of water remaining in each of the microcosms was measured and then filtered through a 47-mm GF/C filter (nominal pore size 1.2 μm). Filters were placed in Petri dishes, wrapped in foil, and frozen for subsequent fluorometric Chl *a* analysis as described above.

Microcosm experiment, September 1992

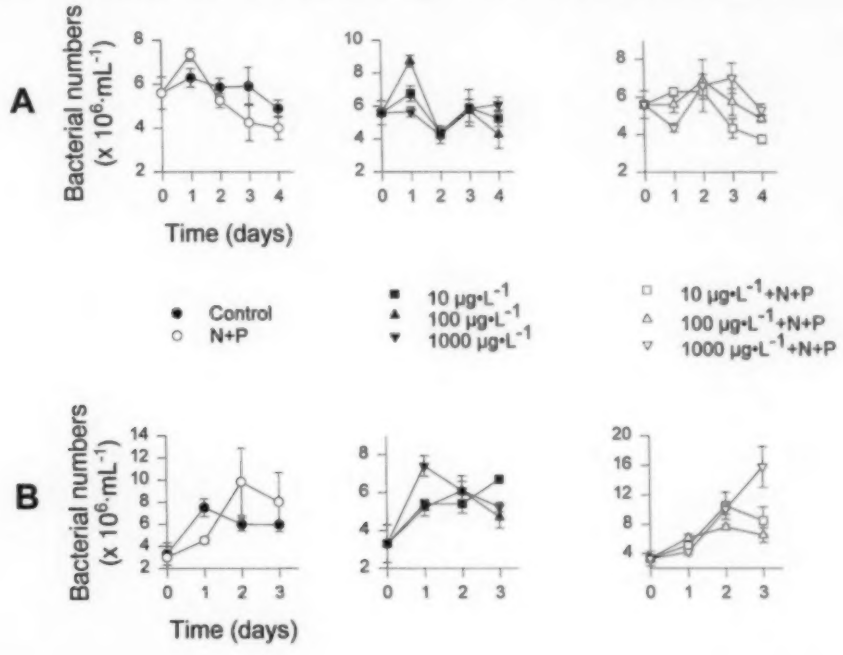
An identical herbicide and nutrient microcosm experiment was conducted in September 1992, with three exceptions. In addition to prescreening the water through a 153-μm-mesh Nitex screen to remove large zooplankters, the water was also passed through a Whatman 934-AH glass fibre filter (nominal pore size 1.5 μm). This prefiltering served two purposes: the removal of bacteriovores to ensure a direct bacterial growth response and the removal of algae to ensure that bacteria were responding only to the nutrient and herbicide amendments and not indirectly to changes in algal metabolism (Coveney and Wetzel 1992). The flasks were also incubated in the dark to select against autotrophs.

Secondly, before to filtration through the 934-AH filters, aliquots of water from each carboy were taken for Chl *a* analysis and bacterial counts were as described above. The filtrate from the 934-AH filters was also sampled for bacterial numbers and Chl *a* to establish that filtration effectively removed most of the algal population, leaving the bacterial population relatively intact.

Thirdly, the September experiment was run for 3 days only. Close inspection of the results from the August microcosm revealed that in most of the treatments, bacterial activity had decreased sharply by day 3 and levelled off by day 4. It was consequently decided to run this microcosm experiment for only 3 days.

© 1997 NRC Canada

Fig. 1. Changes in bacterial numbers with time in response to nutrient and various herbicide treatments in herbicide–nutrient microcosm experiments, Redberry Lake, (A) August 1992 and (B) September 1992. Data are means of three replicates (±1 SE).



Statistical analysis

A one-way analysis of variance (ANOVA) on the Chl *a*, bacterial numbers, production, and metabolism data followed by an a posteriori Tukeys test was used to test for differences between treatments. The significance level for differences among treatments was $P < 0.05$. In addition, a two-way ANOVA, testing herbicide versus nutrient treatments, was used to test for possible interactive effects at the same significance level ($P < 0.05$).

Results

Microcosm experiment, August 1992

Bacterial numbers at T1 (day 1) in the 100 $\mu g \cdot L^{-1}$ treatment were significantly higher than in the 1000 $\mu g \cdot L^{-1}$ or any of the herbicide plus nutrient treatments (Fig. 1A). Other than this result, there were no sigificant differences in bacterial numbers over the course of this experiment.

The greatest stimulus to [^{14}C]glucose uptake was found in the herbicide plus nutrients treatments (Fig. 2A). At T2 (day 2), rates of [^{14}C]glucose uptake in the nutrient and all of the herbicide + N + P treatments were higher than control values (Fig. 2); at T3 (day 3) and T4 (day 4), rates of glucose uptake in the + 1000 $\mu g \cdot L^{-1}$ + N + P treatment only were higher than controls ($P = 0.001$ and 0.003, respectively).

As with [^{14}C]glucose uptake on a volumetric basis, the largest increase in the rate of [^{14}C]glucose uptake on a per-cell basis was seen in those microcosms that received the herbicide plus nutrient additions (Fig. 2B). In all other treatments, glucose uptake per cell remained slightly above or below control values for the duration of the experiment.

The greatest stimulus to TdR incorporation was seen in the N + P treatment, where incorporation rates reached 273 $pmol \cdot L^{-1} \cdot h^{-1}$ at T2 of the experiment (Fig. 3A), a rate that was significantly greater than the control ($P = 0.004$). There was no difference, however, between control and herbicide + nutrient treatments at any time period. TdR incorporation rates in all of the herbicide only treatments were the lowest seen in this experiment and were always below control values (Fig. 3A). The statistical tests, however, revealed a significant difference between the herbicide and the control treatments at T2 only. The two-way ANOVA revealed significant herbicide–nutrient interactions at T2 ($P = 0.022$) and T3 ($P = 0.036$).

Generally, TdR uptake per cell followed the same pattern as TdR uptake on a volumetric basis (Fig. 3B). Higher rates of uptake in the N + P and 10 $\mu g \cdot L^{-1}$ treatments were more clearly differentiated, however, from the other treatments when expressed on a per-cell basis.

Initial Chl *a* values in the microcosm flasks were 0.84 $\mu g \cdot L^{-1}$ (Fig. 4). Chl *a* values in the flasks that received nitrogen plus phosphorus (N + P), N + P plus 10 μg triallate$\cdot L^{-1}$, and N + P plus 100 μg triallate$\cdot L^{-1}$ (Fig. 4) were significantly higher ($P = 0.001$) than control values. In the 1000 μg triallate$\cdot L^{-1}$ treatment the final Chl *a* concentration was 72% below control values, a difference that was statistically significant ($P = 0.044$). A two-way ANOVA revealed a significant effect of nutrients ($P = 0.001$) and herbicide ($P = 0.002$) but no significant interaction of the two ($P = 0.081$).

Microcosm experiment, September 1992

Filtration of Redberry Lake water through 934-AH filters allowed 74% of the bacteria to pass through. Initial bacterial

© 1997 NRC Canada

Fig. 2. [^{14}C]glucose uptake rates on a (A) volumetric and (B) per-cell basis in response to nutrient and various herbicide treatments in herbicide–nutrient microcosm experiments, Redberry Lake, August 1992. Data are means of three replicates (±1 SE).

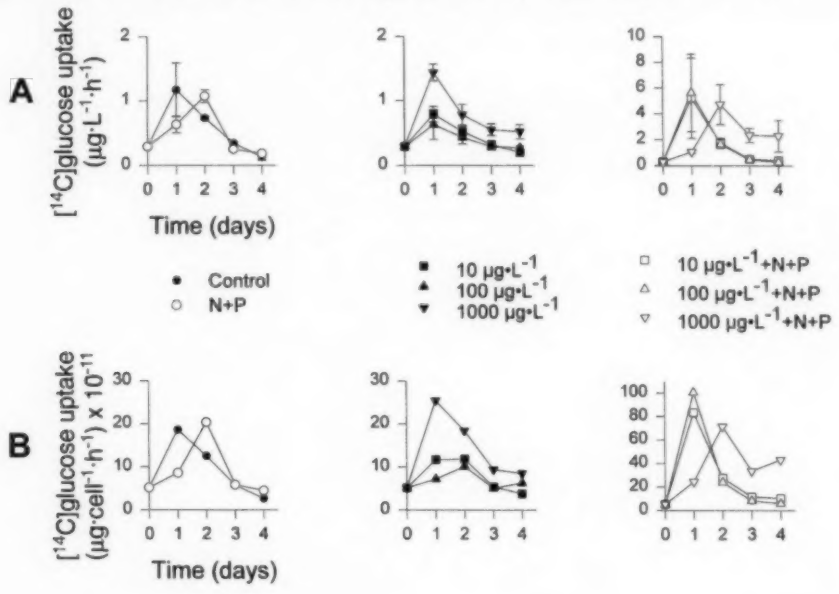
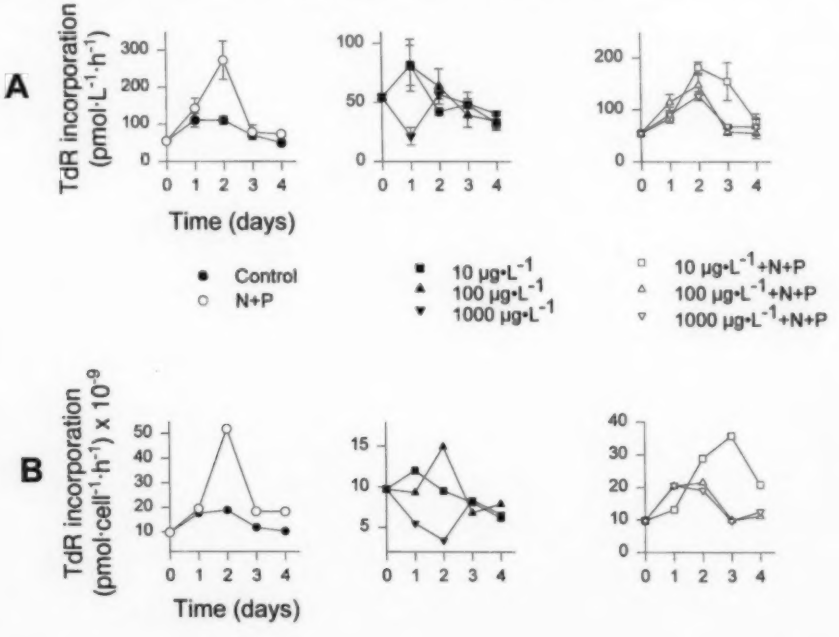


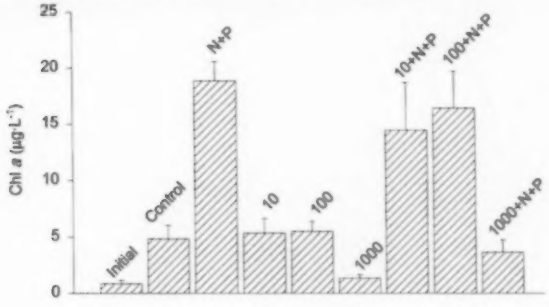
Fig. 3. TdR incorporation rates on a (A) volumetric and (B) per-cell basis in response to nutrient and various herbicide treatments in herbicide–nutrient microcosm experiments, Redberry Lake, August 1992. Data are means of three replicates (±1 SE).



numbers in the microcosms were 3.3×10^6·mL^{-1}. Generally, there was not a great decrease in bacterial numbers in any of the treatments (Fig. 1B), indicating that filtration through the 934-AH filter probably removed most of the grazers. Bacterial numbers in the 1000 µg·L^{-1} + N + P treatments at all sampling times were significantly higher than control numbers (Fig. 1B), and bacterial numbers in the N + P treatment were higher than controls ($P = 0.046$), but at T1 only.

© 1997 NRC Canada

Fig. 4. Initial and final Chl *a* concentrations from the microcosm flasks, Redberry Lake microcosm experiments, August 1992.



The herbicide plus nutrient treatments demonstrated the highest rates of [^{14}C]glucose uptake in this experiment (Fig. 5A). Rates in the 1000 $\mu g \cdot L^{-1}$ + N + P treatments were significantly higher than control or nutrient treatments, but at T2 only. A two-way ANOVA revealed a significant herbicide–nutrient interaction at this time ($P = 0.019$).

Rates of glucose uptake for all treatments, expressed on a per-cell basis, showed the same pattern as glucose uptake on a volumetric basis (Fig. 5B).

As with [^{14}C]glucose rates, rates of thymidine incorporation were highest in the herbicide plus nutrient treatments (Fig. 6A). When these rates were compared with those in the control treatments, they were significantly higher at T2 and T3. Statistically, TdR incorporation rates in the nutrient treatments were significantly higher than the control ($P = 0.014$) or the herbicide treatments ($P = 0.01$), but only at T1 (Fig. 6A). At T2 and T3, there was a significant ($P = 0.001$) nutrient–herbicide interaction.

TdR incorporation for all treatments expressed on a per-cell basis indicated the same trends as volumetric TdR incorporation (Fig. 6B).

Filtration of Redberry Lake water through 934-AH filters removed 77% of Chl *a*. While initial Chl *a* levels in all treatments were 0.40 $\mu g \cdot L^{-1}$, these levels declined over time, indicating that incubation in the dark successfully selected against autotrophs.

Discussion

Nutrient and herbicide effects

A most striking result of this study is that triallate clearly stimulated bacteria, but only when nitrogen and phosphorus were added. Rates of [^{14}C]glucose uptake and TdR incorporation and, in some cases, bacterial numbers were stimulated well above control levels (Figs. 2, 3, 5, and 6) in both the August and September experiments. Amending Redberry Lake water with nutrients and herbicide had a synergistic effect on bacteria, activating measures of bacterial production and growth — an activation that was seen ephemerally when nutrients were added and not at all with herbicide only addition. Synergistic effects of this type have been described in the literature (Coveney and Wetzel 1992; Bell et al. 1993). Bell et al. (1993), for example, found a sensitive balance between organic carbon and nutrient limitation in an oligotrophic, slightly acidified lake in Sweden. Although working in mesocosm enclosures that were located in the lake, their data are similar to data presented here. Thymidine incorporation in Bell et al.'s (1993) study was greatest in mesocosms that were treated with glucose, nitrogen, and phosphorus as compared with either single additions of nutrients and glucose or the control. The response of bacteria was ascribed not only to nutrients, but also to the presence of a labile carbon source as well. Although we lack specific information on actual degradation of triallate by bacteria in Redberry Lake, it seems plausible that bacteria are using the herbicide as a carbon source, but only when nutrients are in sufficient supply. The low molecular weight (304.5) and C:N ratio (9:1) of triallate would certainly make it a suitable carbon substrate for bacterial uptake.

Redberry Lake is totally surrounded by cultivated land that slopes gently on all sides towards the lake. Two major inflows, Oscar and Marshy creeks, drain this cultivated land and bring substantial amounts of water into the lake during spring runoff. Triallate and nutrient addition to the lake will likely be greatest during spring runoff events because mean precipitation in autumn is lower (Atmospheric Environment Service 1986). Based on the results of this study, it would seem that in spring, as long as temperature was not a limiting factor, presence of both triallate and nutrients could stimulate bacterial production and growth in Redberry Lake. At this time of the year, the microbial community of Redberry Lake shows strong signs of nutrient sufficiency (Waiser and Robarts 1995). During the rest of the year, however, this community is severely limited by nitrogen and phosphorus (Waiser and Robarts 1995) and therefore it seems highly unlikely that any triallate entering Redberry Lake, through either aeolian deposition or spray drift, would stimulate bacterial production and growth, unless accompanied by an addition of nutrients. If bacteria cannot use this herbicide as a carbon source, it is possible that triallate may bioaccumulate in the system.

Hydrophobic contaminants, like triallate, that have a half-life of several days in water (Environment Canada 1992) will adsorb to and concentrate in particulate material (plankton) within an aquatic ecosystem if they are not degraded. Lakes with more plankton biomass will tend to have less contaminant per unit biomass and, conversely, oligotrophic lakes should have more (Taylor et al. 1991). In an oligotrophic lake such as Redberry, bioaccumulation of the herbicide may take place by zooplankton grazing on algae which have adsorbed or concentrated high concentrations of herbicide. Additionally, triallate may be strongly adsorbed onto sediment surfaces where foraging, sediment-dwelling amphipods such as *Gammarus* spp. and *Hyallela* spp. could be exposed to high concentrations of this compound (Arts et al. 1995). Bioaccumulation of this lipophilic contaminant into amphipod tissues could theoretically ensue. In fact, following exposure to food containing ^{14}C-labelled triallate, autoradiography of Redberry Lake amphipods revealed that ingested herbicide rapidly became associated with triacyl-rich storage droplets (Arts et al. 1995).

Redberry Lake, however, has very high concentrations of dissolved organic carbon (37 $mg \cdot L^{-1}$). It is well known that DOC can form stable complexes with hydrophobic contaminants and trace inorganic metals (McCarthy and Jiminez 1985). The relatively water-insoluble contaminant is held in solution by partitioning into the more water-soluble DOC.

© 1997 NRC Canada

Fig. 5. [^{14}C]glucose uptake rates on a (A) volumetric and (B) per-cell basis in response to nutrient and various herbicide treatments in herbicide–nutrient microcosm experiments, Redberry Lake, September 1992. Data are means of three replicates (±1 SE).

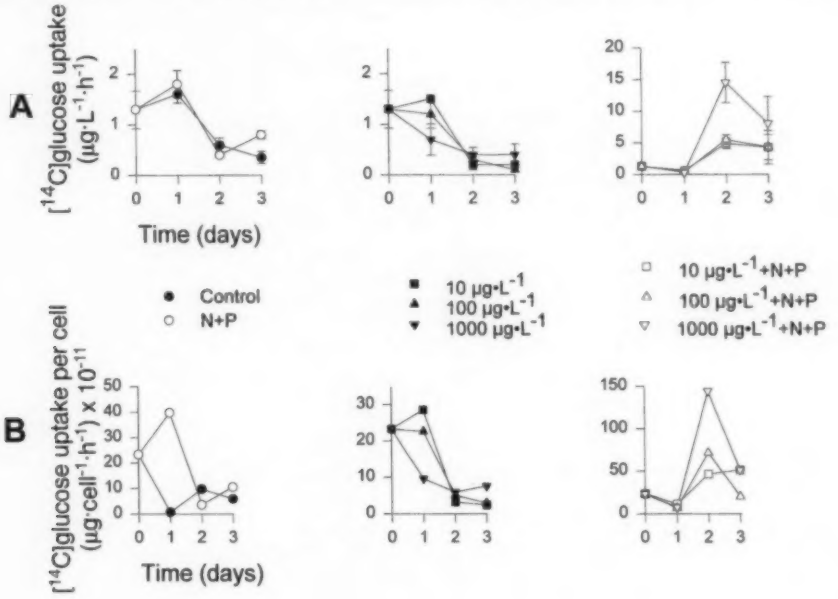
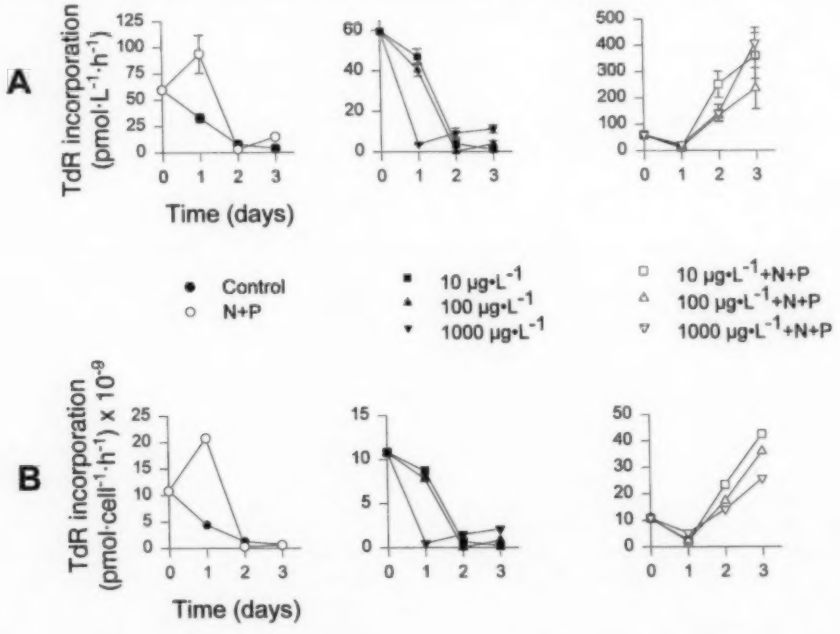


Fig. 6. TdR incorporation rates on a (A) volumetric and (B) per-cell basis in response to nutrient and various herbicide treatments in herbicide–nutrient microcosm experiments, Redberry Lake, September 1992. Data are means of three replicates (±1 SE).



Such partitioning effectively decreases the bioavailabilty of the contaminant mainly because the resulting complex is too large to penetrate biological membranes (Day 1991). The affinity for binding of herbicide to DOC, however, will depend on the size, polarity, and molecular configuration of the DOC, which in turn is dependent on pH and ionic composition of the lake (McKnight 1990; Day 1991).

In Redberry Lake, more research is needed to ascertain

© 1997 NRC Canada

whether triallate adsorbs to plankton or DOC and how this ultimately affects its fate.

Nutrient effects

Addition of nitrogen and phosphorus had an obvious effect on Redberry Lake phytoplankton and bacteria. In the August microcosm experiment, for example, nutrient addition increased Chl *a* levels significantly above control values. While many studies have shown that nutrient additions (N + P) stimulate phytoplankton growth in aquatic ecosystems (Elser et al. 1990), fewer have examined the effects of nutrients on aquatic bacteria. Although temperature and the supply of dissolved organic matter are known to limit bacterial growth (Kirchman 1990; White et al. 1991), it has recently been shown that the supply of dissolved inorganic phosphorus can limit bacterial growth as well (Toolan et al. 1991; Coveney and Wetzel 1992). Nitrogen and phosphorus addition in our microcosm experiments prompted a significant increase in bacterial TdR incorporation (Figs. 3 and 6), but bacterial numbers increased only in the September experiment (Fig. 1) where water was prescreened and then filtered through filters with a nominal pore size of 1.5 μm. Prescreening water through 153-μm pore-size Nitex mesh in the August experiment, however, may not have been enough to exclude ciliates and flagellates (about 2–20 μm, Pernie et al. 1990). This lack of exclusion may account for the decrease in bacterial numbers in the August microcosm experiment.

If phytoplankton and bacteria in this lake are indeed limited by nutrients, one would expect to see other indications of limitation as well. These would include positive responses of phytoplankton to nitrogen and phosphorus additions in nutrient enrichment bioassays, rapid ^{32}P uptake and turnover time throughout the ice-free season (the majority of which is associated with bacteria), and production of high levels of alkaline phosphatase. All of these physiological responses have been recorded for the microbial community of Redberry Lake (Waiser and Robarts 1995) and provide strong support for our microcosm observations of microbial nutrient limitation.

If phosphorus, originating as fertilizer, enters this lake through processes of runoff or aeolian deposition, it will likely be taken up and quickly cycled by the bacterial community. Such domination of inorganic phosphorus uptake by bacteria may have profound effects on the size and species composition of the phytoplankton (Kirchman 1994), which may ultimately affect carbon and energy turnover in this ecosystem.

Herbicide effects

Herbicides may affect aquatic phytoplankton in several possible ways: decreases in measures of algal biomass as measured by Chl *a* or ash-free dry mass (De Noyelles et al. 1982; Kasai and Hanazato 1995), changes in algal growth rate as measured by variations in Chl *a* over time (Abou-Waly et al. 1991), changes in community structure (De Noyelles et al. 1982; Kasai and Hanazato 1995), changes in phytoplankton morphology (Abou-Waly and Shabana 1993), inhibition of photosynthesis (De Noyelles et al. 1982; Kasai and Hanazato 1995; Brown and Lean 1995), and inhibition of nitrogen and phosphorus uptake (Brown and Lean 1995). Data from the August microcosm experiment demonstrated quite clearly that triallate will cause a significant decrease in algal biomass as measured by Chl *a*, but only at concentrations $\geq 1000\ \mu g \cdot L^{-1}$ (Fig. 4). At lower doses (10 and 100 $\mu g \cdot L^{-1}$), there was no significant effect. Kasai and Hanazato (1995) noted a similar direct phytoplankton response when the herbicide simetryn was added to experimental ponds. At low simetryn doses (60–80 $\mu g \cdot L^{-1}$), most phytoplankton individuals withstood negative herbicide effects. Johnson (1986), in a microcosm approach similar to the one used here, found that triallate concentrations >10 $\mu g \cdot L^{-1}$ reduced algal growth (expressed as milligrams of dry weight) by 40%.

The direct effect of high triallate additions will be a decline in algal biomass. Of interest as well, however, are the indirect effects that this decline in algal biomass could have on zooplankton. Although indirect effects were not examined in this investigation, other studies have shown that instability and unexpected events within the food web may result from herbicide addition (De Noyelles 1982; Lampert et al. 1989; Kasai and Hanazato 1995). In all of these studies, a herbicide-induced decline in Chl *a* levels caused a concomitant reduction in zooplankton growth and reproduction. In addition, Johnson (1986) showed that triallate at 100 and 1000 $\mu g \cdot L^{-1}$ reduced algal growth in wetland microcosms by more than 40% at the end of a 30-day period. None of the *Daphnia* introduced into these microcosms survived. In Redberry Lake where *Daphnia* have been nudged into a subsistence niche by increases in lake salinity (Arts et al. 1993), any reduction in food supply, especially at certain times of the year, may push the energy balance of *Daphnia* below the maintenance threshold.

Triallate concentrations reaching 1000 $\mu g \cdot L^{-1}$ in Redberry Lake would only arise as a result of direct overspray or from spring runoff events, and therefore, declines in algal biomass would only be expected in spring. Current summer concentrations of triallate in the lake are <10 $\mu g \cdot L^{-1}$ (M.T. Arts, unpublished data), and at these levels, it would appear that the phytoplankton community will be unaffected, at least for the summer months.

Data on the effects of herbicides in general, and triallate in particular, on aquatic bacterial populations in aquatic ecosystems are scarce. Early work (e.g., Cragg and Fry 1984) concentrated on the effects of herbicides on bacterial numbers. Unfortunately, counts were made by plating bacteria onto solid media, a technique that has been shown to consistently underestimate bacterial numbers. Johnson (1986) tested the effect of the same concentrations of triallate used in the present study on microbial activity (dehydrogenase and phosphatase activity, oxygen consumption, and [^{14}C]glucose mineralization) in simulated prairie wetland microcosms. The results were similar to those presented here (Figs. 1, 2, 3, 5, and 6). Triallate apparently had no effect, positive or negative, on measures of microbial numbers or metabolism.

Conclusion

Three major conclusions may be drawn from this research. The pre-eminent conclusion is that during spring runoff events, nitrogen, phosphorus, and triallate that enter Redberry Lake as a result of cultivation practices in the basin have the potential to positively stimulate bacterial growth in the lake. This increase, which may occur because bacteria are using the herbicide as a carbon source, only arises when nitrogen and phosphorus are present and available. In summer, when the microbial population is nutrient limited, no such activation

© 1997 NRC Canada

will occur, and therefore the possibility exists that triallate could biaccumulate, although the presence of high concentrations of DOC in the lake might influence this process. Secondly, triallate at high enough concentrations will significantly depress algal growth. And thirdly, stimulation of algal and bacterial growth will occur when nutrients, originating from fertilization of arable land, enter the lake ecosystem. Changes in carbon and energy cycling within the ecosystem could theoretically ensue as a result of any of these changes.

Acknowledgements

We thank Vijay Tumber for field and laboratory support and two anonymous reviewers for comments that improved the style and content of the manuscript. We also thank Monsanto Corporation, St. Louis, Mo., for providing technical advice.

References

Abou-Waly, H., and Shabana, E.F. 1993. Recovery of *Nostoc muscorum* previously exposed to some triazine and phenylurea herbicides. Bull. Environ. Contam. Toxicol. **50**: 665–675.

Abou-Waly, H., Abou-Setta, M.M., Nigg, H.N., and Mallory, L.I. 1991. Growth response of freshwater algae, *Anabaena flos-aquae*, and *Selenastrum capricornutum* to atrazine and hexazinone herbicides. Bull. Environ. Contam. Toxicol. **46**: 223–229.

Arts, M.T., Robarts, R.D., and Evans, M.S. 1993. Energy reserve lipids of zooplanktonic crustaceans from an oligotrophic lake in relation to food resources and temperature. Can. J. Fish. Aquat. Sci. **50**: 2404–2420.

Arts, M.T., Ferguson, M.F., Glozier, N.E., Robarts, R.D., and Donald, D.B. 1995. Spatial and temporal variability in lipid dynamics of common amphipods: assessing the potential for uptake of lipophilic contaminants. Ecotoxicology, **4**: 91–113.

Atmospheric Environment Service. 1986. Climatic atlas of Canada. Map series No. 2. Precipitation. Canadian Government Publishing Centre, Ottawa, Ont.

Bell, R.T., Vrede, K., Stensdotter-Blomberg, U., and Blomqvist, U. 1993. Stimulation of the microbial food web in an oligotrophic, slightly acidified lake. Limnol. Oceanogr. **38**: 1532–1538.

Brown, L.S., and Lean, D.R.S. 1995. Toxicity of selected pesticides to lake phytoplankton measured using photosynthetic inhibition compared to maximal uptake rates. Environ. Toxicol. Chem. **14**: 93–98.

Cole, J.J., Findlay, S., and Pace, M.L. 1988. Bacterial production in fresh and saltwater ecosystems: a cross system overview. Mar. Ecol. Prog. Ser. **43**: 1–10.

Coveney, M.F., and Wetzel, R.G. 1992. Effects of nutrients on specific growth rate of bacterioplankton in oligotrophic lake water. Appl. Environ. Microbiol. **58**: 150–156.

Cragg, B.A., and Fry, J.C. 1984. The use of microcosms to simulate field experiments to determine the effects of herbicides on aquatic bacteria. J. Gen. Microbiol. **130**: 2309–2316.

Day, K. 1991. Effects of DOC on accumulation and acute toxicity of fenvalerate, deltamethrin and cyhalothrin to *Daphnia magna* (Strauss). Environ. Toxicol. Chem. **10**: 91–101.

De Noyelles, F., Kettle, W.D., and Sinn, D.E. 1982. The response of plankton communities in experimental ponds to atrazine, the most heavily used pesticide in the United States. Ecology, **63**: 1285–1293.

Donald, D.B., and Syrgiannis, J. 1995. Occurrence of pesticides in prairie lakes in Saskatchewan in relation to drought and salinity. J. Environ. Qual. **24**: 266–270.

Elser, J.J., Marzolf, E.R., and Goldman, C.R. 1990. Phosphorus and nitrogen limitation of phytoplankton growth in freshwaters of North America: a review and critique of experimental enrichments. Can. J. Fish. Aquat. Sci. **47**: 1468–1477.

Environment Canada. 1992. Canadian water quality guidelines for triallate. Scientific Format Series. Environmental Quality Guideline Division, Water Quality Branch, Inland Waters Directorate, Environment Canada, Ottawa, Ont.

Giesy, J.P., Jr., and Allred, P.M. 1985. Replicability of aquatic multispecies test systems. *In* Multispecies toxicity testing. *Edited by* John Cairns, Jr. Pergamon Press Inc., Tarrytown, N.Y. pp. 187–250.

Grover, R., Kerr, L.A., Bowren, K.E., and Khan, S.U. 1988. Airborne residues of triallate and trifluralin in Saskatchewan. Bull. Environ. Contam. Toxicol. **40**: 683–688.

Hobbie J.E., and Crawford, C.C. 1969. Respiration correction for bacterial uptake of dissolved organic compounds in natural waters. Limnol. Oceanogr. **14**: 528–532.

Humburg, N.E. 1989. Herbicide handbook of the Weed Science Society of North America. 6th ed. Weed Science Society of America, Champaign, Ill.

Hurlbert, S.H. 1984. Pseudoreplication and the design of ecological field experiments. Ecol. Monogr. **54**: 187–211.

Johnson, B.T. 1986. Potential impact of selected agricultural chemical contaminants on a northern prairie wetland: a microcosm evaluation. Environ. Toxicol. Chem. **5**: 473–485.

Kasai, F., and Hanazato, T. 1995. Effects of the triazine herbicide, simetryn, on freshwater plankton communities in experimental ponds. Environ. Pollut. **89**: 197–202.

Kirchman, D.L. 1990. Limitation of bacterial growth by dissolved organic matter in the subarctic Pacific. Mar. Ecol. Prog. Ser. **62**: 47–54.

Kirchman, D.L. 1994. The uptake of inorganic nutrients by heterotrophic bacteria. Microb. Ecol. **28**: 255–271.

Lampert, W., Fleckner, W., Pott, E., Schober, U., and Storkel, K. 1989. Herbicide effects on planktonic systems of different complexity. Hydrobiologia, **188–189**: 415–424.

Lewis, E.T. 1991. Herbicides used for agricultural weed control in western Canada, 1987–89. Agriculture Statistics, Economics Branch, Manitoba Department of Agriculture, Winnipeg, Man.

McCarthy, J.F., and Jiminez, B.D. 1985. Reduction in bioavailability to bluegills of polycyclic aromatic hydrocarbons bound to dissolved humic material. Environ. Toxicol. Chem. **4**: 511–521.

McKnight, D.M. 1990. How do organic acids interact with solutes, surfaces and organisms? *In* Organic acids in aquatic ecosystems. *Edited by* E.M. Perdue and E.T. Gjessing. John Wiley & Sons, Inc., New York. pp. 223–243.

Mitsch, W.J., and Gosselink, J.G. 1993. Wetlands. 2nd ed. Van Nostrand Reinhold Company Inc., New York.

Nusch, E.A. 1980. Comparison of different methods for chlorophyll and pheopigment analysis. Ergeb. Limnol. **14**: 14–36

Pernie, G.I., Scavia, D., Pace, M.I., and Carrick, H.J. 1990. Micrograzer impact and substrate limitation of bacterioplankton in Lake Michigan. Can. J. Fish. Aquat. Sci. **47**: 1836–1841.

Pomroy, A.J. 1984. Direct counting of bacteria preserved with Lugol's iodine solution. Appl. Environ. Microbiol. **47**: 1191–1192.

Porter, K.G., and Feig, Y. 1980. The use of DAPI for identifying and counting aquatic microflora. Limnol. Oceanogr. **25**: 943–948.

Robarts, R.D., and Wicks, R.J. 1989. [Methyl-^{3}H]thymidine macromolecular incorporation and lipid labelling: their significance to DNA labelling during measurements of aquatic bacterial growth rate. Limnol. Oceanogr. **34**: 213–222.

Robarts, R.D., and Zohary, T. 1993. Fact or fiction — bacterial growth rates and production as determined by [methyl-^{3}H]-thymidine. Adv. Microb. Ecol. **13**: 371–425.

© 1997 NRC Canada

Robarts, R.D., Evans, M.S., and Arts, M.T. 1992. Light, nutrients and water temperatures as determinants of phytoplankton production in two saline prairie lakes with high sulphate concentrations. Can. J. Fish. Aquat. Sci. **49**: 2281–2290.

Simon, M., Cho, B.C., and Azam, F. 1992. Significance of bacterial biomass in lakes and the ocean: comparison to phytoplankton biomass and biogeochemical implications. Mar. Ecol. Prog. Ser. **86**: 103–110.

Taylor, W.D., Carey, J.H., Lean, D.R.S., and McQueen, D.J. 1991. Organochlorine concentrations in the plankton of lakes in southern Ontario and their relationship to plankton biomass. Can. J. Fish. Aquat. Sci. **48**: 1960–1966.

Toolan, T.,Wehr, J.D., and Findlay, S. 1991. Inorganic phosphorus stimulation of bacterioplankton production in a mesoeutrophic lake. Appl. Environ. Microbiol. **57**: 2074–2078.

Van Donk, E., Prins, H., Voogd, H.M., Crum, S.J.H., and Brock, T.C.M. 1995. Effects of nutrient loading and insecticide application on the ecology of *Elodea*-dominated freshwater microcosms. 1. Responses of plankton and zooplanktivorous insects. Arch. Hydrobiol. **133**: 417–439.

Waiser, M.J., and Robarts, R.D. 1995. Microbial nutrient limitation in prairie saline lakes with high sulfate concentration. Limnol. Oceanogr. **40**: 566–574.

Waite, D.T., Sommerstad, H., Grover, R., and Kerr, L.A. 1986. South Saskatchewan Water Study (1985 interim report). Environmental Protection Service, Western and Northern Region, W&NR 86/87-CP(EPS)-3.

Waite, D.T., Grover, R., Westcott, N.D., Irvine, D.G., Sommerstad, H., and Kerr, L.A. 1992. Pesticides in groundwater, surface water and spring runoff in a small Saskatchewan watershed. Environ. Toxicol. Chem. **11**: 741–748.

Waite, D.T., Grover, R., Westcott, N.D., Irvine, D.G., Kerr, L.A., and Sommerstad, H. 1995. Atmospheric deposition of pesticides in a small Saskatchewan watershed. Environ. Toxicol. Chem. **14**: 1171–1175.

White, P.A., Kalff, J., Rasmussen, J.B., and Gasol, J.M. 1991. The effect of temperature and algal biomass on bacterial production and specific growth rate in freshwater and marine habitats. Microb. Ecol. **21**: 99–118.

Williamson, D.A. 1984. A preliminary investigation into the presence of agricultural pesticides in the La Salle and Assiniboine rivers, Manitoba, Canada. Environmental Management Services Branch, Manitoba Department of Environment and Workplace Safety and Health, Water Standards and Studies Report No. 84-5.

Wright, R.T. 1978. Measurement and significance of specific activity in the heterotrophic bacteria of natural waters. Appl. Environ. Microbiol. **36**: 297–305.

© 1997 NRC Canada

Physical and chemical limnology of 59 lakes located between the southern Yukon and the Tuktoyaktuk Peninsula, Northwest Territories (Canada)

Reinhard Pienitz, John P. Smol, and David R.S. Lean

Abstract: Water chemistry and other limnological data gathered for 59 lakes in the Yukon and the adjacent Northwest Territories (Canada) were interpreted using linear regression and principal components analysis. The study sites represent lakes from a wide range of ecoclimatic regions, spanning large latitudinal (60°37′–69°35′N) and altitudinal gradients (15–1387 m above sea level). Water samples collected from each lake were analysed for concentrations of major ions, trace metals, nutrients, and chlorophyll *a*. Most of the lakes were dilute (mean conductivity = 160 $\mu S \cdot cm^{-1}$) and slightly acidic to alkaline (pH range = 5.9–9.3). Their ionic composition varied from Ca–Cl–Na waters near the Arctic Ocean to Ca–HCO_3 waters further inland, reflecting differences in local drainage basins and proximity to the sea. Arctic and alpine sites generally showed many similarities, but considerable differences in water chemistry were observed among sites in the interior of the Yukon Territory. These can be related mainly to differences in bedrock geology and catchment vegetation.

Résumé : Des données sur les caractéristiques limnologiques et chimiques obtenues pour 59 lacs dans le Yukon et la partie adjacente des Territoires du Nord-Ouest (Canada) ont été interprétées à l'aide de l'analyse de régression linéaire et de l'analyse des composantes principales. Les sites d'étude sont des lacs qui représentent une grande variété de régions écoclimatiques, échelonnées sur de longs gradients de latitude (60°37′–69°35′N) et d'altitude (15 à 1387 m au-dessus du niveau de la mer). Les échantillons d'eau prélevés dans chacun des lacs ont été analysés pour déterminer la concentration des ions traces, des métaux traces, des éléments nutritifs et de la chlorophylle *a*. La plupart des lacs étaient pauvres en solutés (conductivité moyenne de 160 $\mu S \cdot cm^{-1}$) et leur pH variait de légèrement acide à alcalin (étendue de pH = 5,9 à 9,3). La composition ionique a varié de Ca–Cl–Na près de l'océan Arctique à Ca–HCO_3 vers l'intérieur des terres, ce qui reflète des différences dans les bassins hydrographiques locaux et la proximité de la mer. De manière générale, les sites arctiques et alpins ont présenté de nombreuses similitudes, mais des différences considérables en ce qui a trait à la chimie de l'eau ont été observées entre les sites situés à l'intérieur du Territoire du Yukon. Ces différences peuvent être liées aux différences touchant les caractéristiques du substrat rocheux et de la végétation du bassin versant.
[Traduit par la Rédaction]

Introduction

Current general circulation models predict increased global temperatures, with projected greenhouse warming greater in northern latitudes (e.g., Luckman 1989; Roots 1989; Boer et al. 1990). For example, the CO_2-doubling scenarios of Manabe and Wetherald (1986) show summertime temperature increases of 7°C and soil moisture decreases of 50% for the subarctic area bordering on the west coast of Hudson Bay. The latter estimate represents the most extreme change in North America (Bello and Smith 1990). Ecosystems in high-latitude regions are expected to be particularly sensitive to a given change in magnitude and timing of available energy and to changes in physical and geochemical conditions (Roots 1989; Maxwell 1992). Climatic changes of this order would therefore have serious implications for energy-balance processes and hydrological budgets of northern lakes.

If greenhouse warming occurs, the chemistry of freshwater ecosystems will be greatly affected through, for example, changes in precipitation/evaporation ratios and runoff, duration of ice cover, thickness of snow cover, and forest fires in the catchments (Schindler et al. 1990). Physical and chemical cycles of lakes would be altered in fundamental ways, which in turn govern lake productivity and decomposition processes. Some preliminary work considers the consequences for individual aspects of lakes, such as the effect of flooding on the nutrients of lakes in the Mackenzie Delta (Lesack et al. 1991). However, effects on the entire ecosystem are unknown and unpredictable at present. The greatest limitation to previous studies is that they focused on single problems or issues, including a very limited number of environmental variables.

Received October 11, 1995. Accepted July 31, 1996.
J13105

R. Pienitz.[1] Centre d'études nordiques et département de géographie, Université Laval, Québec, QC G1K 7P4, Canada.
J.P. Smol. Paleoecological Environmental Assessment and Research Laboratory (PEARL), Department of Biology, Queen's University, Kingston, ON K7L 3N6, Canada.
D.R.S. Lean. Canada Centre for Inland Waters, National Water Research Institute, Box 5050, Burlington, ON L7R 4A6, Canada.

[1] Author to whom all correspondence should be addressed.
e-mail: reinhard.pienitz@cen.ulaval.ca

© 1997 NRC Canada

Variable by variable investigations, in which each gradient is treated separately, effectively represent the data, but do not show interrelationships either between gradients or between species (Neilson and Stevens 1987). Thus, they fail to integrate factors that structure freshwater ecosystems.

Northern lakes have traditionally received less attention from climatologists than the vegetated landscape, perhaps because they are ecosystems in which it is difficult to set up and maintain sensitive equipment. In addition, lakes in many northern regions are shallow and typically surrounded by permafrost, like those in the Yellowknife area and on the Tuktoyaktuk Peninsula. This will enhance the role of lakes in regional water balances of northern landscapes, since these water bodies are more likely to become ephemeral, which carries biotic as well as hydrologic and climatologic implications (Bello and Smith 1990; Kling et al. 1991). In the context of climate change, improved understanding of processes related to lake evaporation will become increasingly important in subarctic and arctic regions.

The most recent general reviews of arctic limnology are by Hobbie (1980, 1984), Hammar (1989), and Vincent and Ellis-Evans (1989). Since then, a number of agencies and research groups carried out surveys or reviews on water chemistry of freshwater lakes and ponds in high-latitude regions, including the Yukon Territory, adjacent Alaska, and the Northwest Territories (e.g., Lindsey et al. 1981; Ennis et al. 1982; Horler et al. 1983; Shortreed and Stockner 1986; Welch and Legault 1986; Fee et al. 1988; Anema et al. 1990*a*, 1990*b*; Kling et al. 1992; Eilers et al. 1993). Among these investigations, several were targeted at specific questions such as assessments of fisheries resources. The latter were therefore usually limited to relatively small geographic areas and could not address the effects of climatic and vegetational changes over large environmental gradients. In addition, large areas in the western Arctic of North America remain unexplored limnologically.

In this paper, we interpret the data we have gathered from a limnological survey of 59 lakes located between Whitehorse in the Yukon and Tuktoyaktuk in the Northwest Territories (Fig. 1). Our study sites cross several ecoclimatic regions from boreal forest in the south to arctic tundra in the north. This investigation is part of a more extensive study of algal distributions (diatoms and chrysophytes) and their relationship to environmental gradients from a large number of sites in northern Canada and Scandinavia (Pienitz 1993; Pienitz et al. 1995*a*, 1995*b*).

The objectives of the present study are to describe the physical and chemical characteristics of our study sites and to compare our results with previously reported data on lakes from subarctic regions. Despite the recognized environmental and ecological importance of freshwater ecosystems in high-latitude regions, very little or no baseline data exist for many sites. Thus, another goal of this study is to collect data that will allow us to define the natural state of the present systems, thereby serving as reference data for future monitoring programs. Such data should be useful as baseline data for the management of freshwater ecosystems in these remote northern areas.

Description of study area

No previous limnological information was available for most of the 59 study lakes. Likewise, many of these lakes are unnamed, and so we numbered them in consecutive order of sampling. Samples were collected during the month of July 1990. The lakes are distributed along a south–north transect between Whitehorse and Tuktoyaktuk (Fig. 1). Between Whitehorse and Inuvik, all lakes were sampled from an inflatable boat whereas sampling of lakes on the Tuktoyaktuk Peninsula (26–55) (Fig. 1, inset) was carried out from a helicopter equipped with pontoons. All lakes studied are natural, with the exception of lake 4, which is artificially dammed.

Regional climate and hydrology

The study region comprises four major ecoclimatic provinces (Cordilleran, Subarctic Cordilleran, Subarctic, Arctic) as recognized by the Ecoregions Working Group (1989). The climate of different regions within the study area is summarized in Table 1. The steep climatic (and altitudinal) gradients along the sampling transect are particularly strong in the treeline region between Inuvik and Tuktoyaktuk, only 130 km apart, where marked physiographic changes (transition from forested to treeless areas) account for drastic changes in surface albedo (Ritchie 1977, 1984). As a result, spring breakup on small lakes (5–50 ha) near Tuktoyaktuk is more than a month later (late June to mid-July) and freeze-up in the fall comes earlier (end of August to early September) (Fee et al. 1988) than at Inuvik. In the northern forest zone, small lakes are ice free by June 15 and freeze over by October 15 whereas large (>50 ha) lakes become ice free roughly 2 weeks later than small ones.

Lakes in the southern part of the Yukon are mostly dimictic with protracted periods of ice cover, usually extending from October–November to May–June (Environment Canada 1985–1990). According to Allen (1964), the mean number of days between the clearing of ice from lakes and initial ice formation is about 90–100 days in the Inuvik area whereas the ice-free period lasts considerably longer (about 150 days) in the Whitehorse area.

Tuktoyaktuk lakes

In the Northwest Territories, we restricted our sampling to upland sites in the Inuvik area and on the Tuktoyaktuk Peninsula. The hydrological regime of these arctic lakes is largely controlled by a combination of low precipitation and permafrost, with snowmelt runoff in the spring being the dominant hydrologic event (Anema et al. 1990*a*). Water balance studies have shown that, in summer, evaporation from lake surfaces (about 230 mm) is generally greater than precipitation (10 mm) (e.g., Sheath and Hellebust 1978; Hobbie 1984; Sheath 1986; Marsh and Bigras 1988; Marsh 1989; Bigras 1990). Seasonal variations in surface water chemistry of arctic lakes are thus related mainly to dilution by snowmelt and runoff and to concentration by evaporation and exclusion from ice and (or) permafrost (cryoconcentration) (e.g., Hobbie 1984; Kling et al. 1992). These processes can cause considerable fluctuations in the chemical concentrations of major ions in arctic lakes (e.g., Howard and Prescott 1973; Schindler et al. 1974; Prentki et al. 1980; Welch and Legault 1986; Cornwell 1992). It is unlikely, however, that such seasonal variations strongly affected the major chemical groupings described below, given that all lakes were sampled during a short time span in July 1990. However, it should be noted that the water

© 1997 NRC Canada

Table 1. Climate data from the study area.

	Whitehorse (60°43′N, 135°04′W)	Mayo (63°36′N, 135°53′W)	Dawson City (64°04′N, 139°26′W)	Inuvik (68°21′N, 133°43′W)	Tuktoyaktuk (69°27′N, 133°02′W)
January mean daily air temperature (°C)	−20.0	−27.0	−29.0	−28.0	−27.0
July mean daily air temperature (°C)	16.0	15.0	16.0	13.0	10.0
Mean annual daily temperature (°C)	−1.3	−4.0	−5.0	−10.0	−11.0
Mean annual precipitation (mm)	268.9	293.0	325.1	266.0	137.6
Snow depth (mm)	145.8	144.4	151.8	175.0	56.0
Growing degree-days above 5°C	897.1	983.0	996.5	654.0	372.0
Mean annual global solar radiation (kly · yr^{-1})[a]	na	na	85.0	78.0	77.0
Mean annual net radiation (kly · yr^{-1})	na	na	20.0	14.0	10.0
Mean maximum lake ice thickness (cm)	na	na	150.0	170.0	180.0

Note: na, not available.
[a]1 ly = 0.70 kW · m^{-2}.

chemistry data we recorded for our 59 lakes represent summer concentrations rather than annual averages.

Vegetation

The major vegetation zones of the study area include boreal forest in the south and arctic tundra in the north, with transitional forest–tundra or woodland in between, and alpine tundra in mountainous and high-elevation areas. Trees cover most of the plateaus and valleys in the south and form closed-canopy forests, depending on site conditions. Tree species include white spruce (*Picea glauca*), black spruce (*Picea mariana*), larch (*Larix laricina* var. *alaskensis*), alpine fir (*Abies lasiocarpa*), lodgepole pine (*Pinus contorta*), aspen (*Populus tremuloides*), balsam poplar (*Populus balsamifera*), and paper birch (*Betula papyrifera* ssp. *humilis*) (Oswald and Senyk 1977). Several species occur near the arctic treeline, but white and black spruce are the most prevalent. Alpine tundra consists of several communities ranging from sedge meadows or tussock fields to lichens growing on rocks.

The vegetation of the forest–tundra zone shows abundant evidence of the effects of natural fires, with upland sites usually dominated by white spruce with an understory of lichens (*Cladina* spp. and *Stereocaulon* spp.) and ericoid shrubs (*Vaccinium uliginosum, Vaccinium vitis-idaea, Ledum decumbens,* and *Oxycoccus* spp.). Poorly drained sites are occupied by *Picea mariana* stands with varied ground cover dominated by *Cladina* spp., *Sphagnum* spp., and *Betula glandulosa*. Upland sites in the tundra zone are dominated by dwarf shrubs and lichens whereas poorly drained peaty areas show ice-wedge polygon and frost hummock development with dominance by *Eriophorum vaginatum* and *Carex* stands (Koivo and Ritchie 1978).

Materials and methods

The lakes were chosen to minimize differences in basin morphometry, and therefore are usually isolated, small- to medium-sized basins (see below), circular in shape, and exceed 1 m in water depth. Almost all the study lakes are located in primary watersheds, receiving no significant drainage from rivers or from other lakes. Because of logistic constraints when sampling from a helicopter, the amount and type of information collected from each lake varied slightly from area to area. Hence, concentrations of oxygen and profiles of temperature were not recorded for all 59 lakes. Epilimnion and thermocline depths could only be measured when sampling from an inflatable boat.

Elevations were taken from topographic maps. The surface areas of all lakes were determined by digitizing from 1 : 50 000 scale maps. Maximum depths were generally recorded during repeated sampling of lake surface sediments and were, in a few cases, based on exploratory sounding transects using a Ray Jeff MX2550 echo sounder. Other general features of the lakes, such as drainage patterns, extent of emergent vegetation, shore characteristics, and catchment vegetation, were determined from on the ground observations and are summarized in Pienitz (1993).

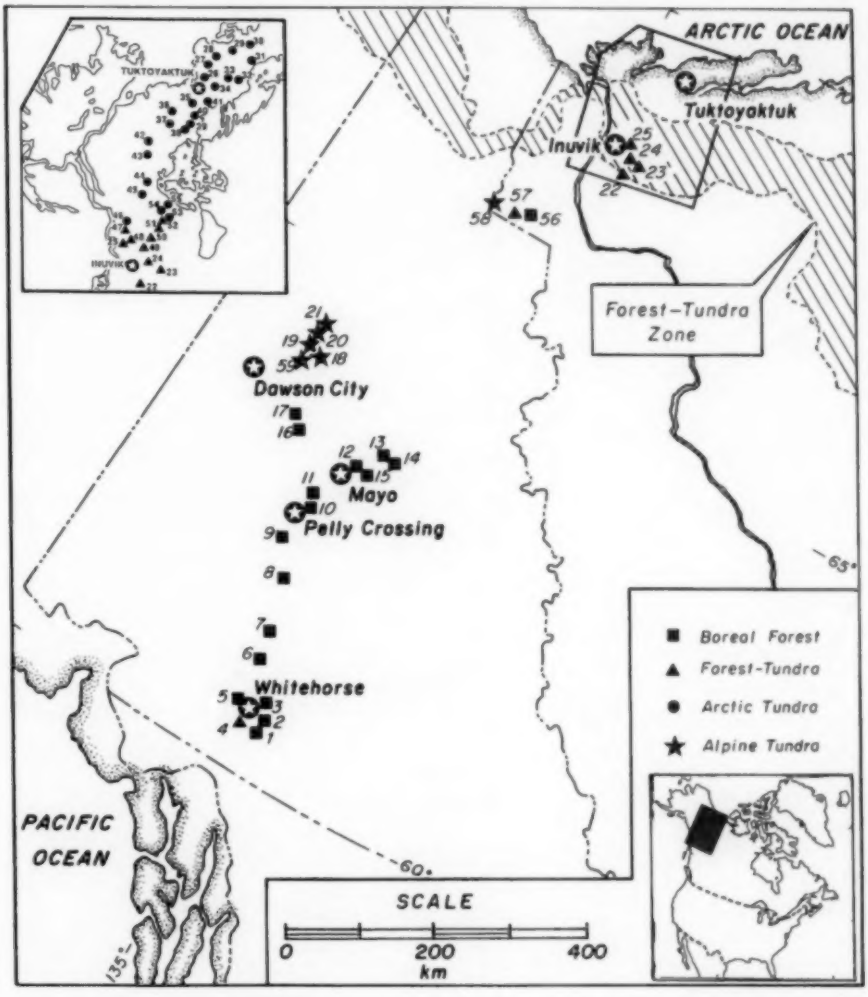
Several statistical approaches were used to describe the patterns observed in the physical, chemical, and environmental variables (see below). The abbreviations in parentheses correspond to the labels used in the figures and tables.

Physical and chemical variables

Limnological field measurements were usually made at one station near the deepest part of the lake, which, because of the simple circular morphometry of most basins, was generally near the centre. A

© 1997 NRC Canada

Fig. 1. Location of the 59 study sites and major towns in the Yukon and Northwest Territories.



standard 22-cm Secchi disk was used to measure water transparency (TRANS). Profile measurements of salinity, conductivity (COND), and water temperature (TEMP) were recorded with a YSI model 33 SCT meter. Salinity measurements were continuously below detection levels, except for two lakes in the vicinity of Whitehorse (1, 2), and are therefore not included in the analyses. The dissolved oxygen content (DO) and pH of the surface water were measured with a YSI model 54 oxygen meter and Hanna field pH meter, respectively.

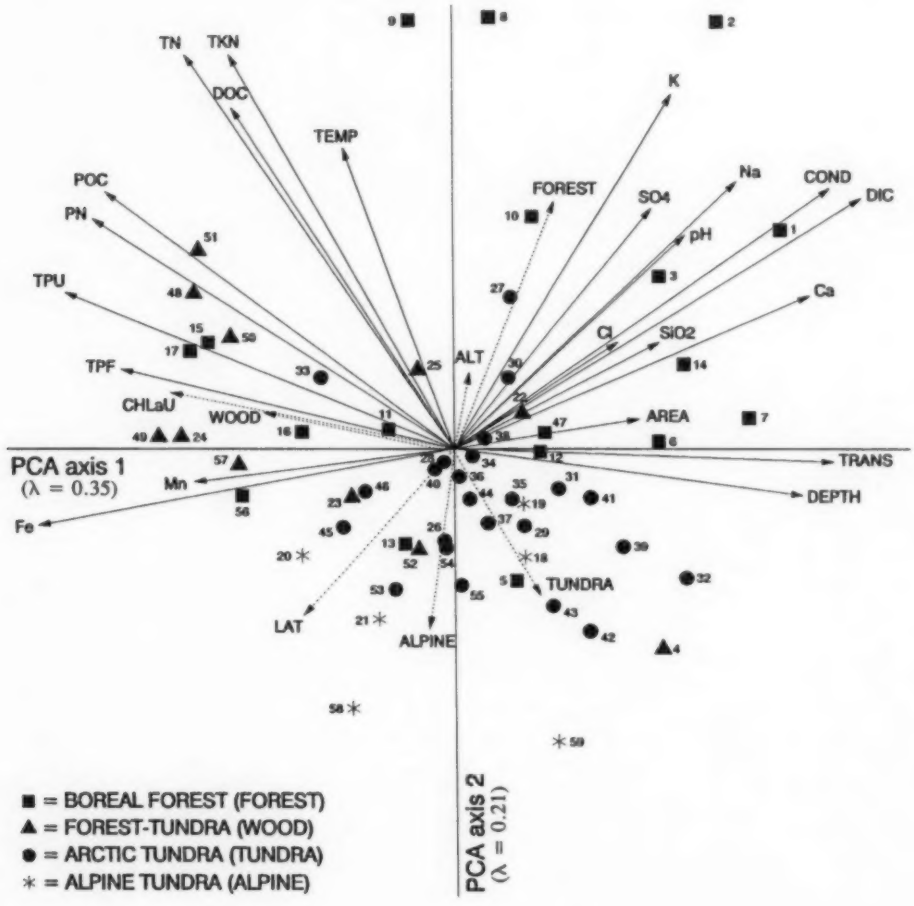
Water samples for chemical analyses were taken from approximately 0.5 m water depth using polyethylene bottles. Samples were immediately treated in the field according to sampling procedures outlined in the *Analytical Methods Manual* (Environment Canada 1979). All samples were kept cold and in the dark before being shipped to the National Water Research Institute (Burlington, Ont.) for additional analyses.

Twenty-three chemical variables were measured on each water sample using standard procedures. Water for nutrient analyses (nitrite (NO_2), nitrate (NO_3), ammonia (NH_3), soluble reactive phosphate–phosphorus (SRP), dissolved organic carbon (DOC), dissolved inorganic carbon (DIC), and total Kjeldahl nitrogen (TKN)) was filtered through 47 mm diameter cellulose acetate filters (pore size = 0.45 μm) and placed in rinsed 100-mL glass bottles. The same filtration was used to obtain water for analyses of total "dissolved" phosphorus (TFP). One millilitre of 30% H_2SO_4 was added to both the filtered and the unfiltered phosphorus fractions (TPU, including phosphorus in particulate and dissolved phase) and placed in 100-mL glass bottles.

Depending on lake productivity, between 100 and 500 mL of water was filtered through ashed 47 mm diameter Whatman GF/F glass microfibre filters (particle retention = 0.7 μm). These filters were stored in plastic petri dishes, wrapped in aluminum foil, and later analysed for particulate organic carbon (POC) and particulate nitrogen (PN). About 1 mL of saturated $MgCO_3$ solution was added to a second 500-mL sample of lake water, which was then also filtered through glass microfibre filters for analyses of total chlorophyll (CHLAU and CHLAC (uncorrected and corrected for phaeophytin)). These filters were folded once, placed in a plastic petri dish, and then wrapped in aluminum foil. Major ions (sodium (Na), potassium (K), calcium (Ca), chloride (Cl), sulphate (SO_4)), dissolved silica (SiO_2) as well as the trace metals iron (Fe) and man-

© 1997 NRC Canada

Fig. 2. PCA of the physical, chemical, and environmental variables in the 59-lake data set. Passive variables are represented by arrows with broken lines.



ganese (Mn) were determined from samples stored in 100-mL plastic bottles. Water samples for analyses of Fe and Mn were preserved with 1 mL of concentrated HNO_3. The *Analytical Methods Manual* (Environment Canada 1979) provides further details concerning analytical methods and sample treatment.

Morphometric and geographic–environmental variables

Lake surface area (AREA), altitude (ALT), maximum depth (DEPTH), latitude (LAT), and longitude (LONG) were determined for each site whereas shortest distance from northern treeline (DIST) was determined for a subset of 31 sites located in the Inuvik and Tuktoyaktuk areas. In addition, the lakes were classified according to the four main vegetation zones: boreal forest (FOREST, including some lakes in peatlands), subarctic woodland (WOOD, includes both forest–tundra and lichen–woodland), arctic tundra (TUNDRA), and alpine tundra (ALPINE) (Fig. 1).

Statistical analyses

Canonical ordination combines the simplicity of regression models with the power of ordination models to detect the internal structure in a data set, including a relatively large number of interrelated variables (Van Tongeren et al. 1992). We therefore chose to use principal components analysis (PCA) to study the trends and the relationships between sites and environmental variables and to help decipher which processes may govern the observed patterns. PCA was also used to screen the data to detect any unusual or outlier sampling sites, which were excluded from further statistical analyses. The variables were tested for skewedness and, if necessary, their data were $\ln(x + 1)$ transformed (Zar 1984) to approximate a normal distribution. PCA analyses were conducted on the $\ln(x + 1)$ transformed data files. All analyses were computed using the program CANOCO, version 3.1 (Ter Braak 1990*a*, 1990*b*).

Groups of significantly ($P \leq 0.05$) correlated variables were identified from a Pearson correlation matrix with Bonferroni-adjusted probabilities (Wilkinson 1988). The correlation matrix, which is based on the $\ln(x + 1)$ transformed data and includes all available data, shows patterns of correlation between variables that are discussed below.

Results and discussion

Physical and geographic–environmental variables

Lakes sampled in the Yukon varied considerably in elevation, ranging from 15 to 1387 m above sea level (asl) (Table 2). Lakes of the Yukon Plateau in the southern Yukon generally ranged in elevation from 580 to 1020 m asl. The five alpine

© 1997 NRC Canada

sites in the South Ogilvie Mountains (18–21, 59) occurred roughly between 1000 and 1400 m asl whereas the alpine site in the Richardson Mountains (58) was located only at 550 m asl, thereby indicating the progressive drop in alpine treeline elevation with increasing latitude. Low-elevation sites, where lakes were generally below 150 m asl, were predominantly regions of forest–tundra and arctic tundra. Lakes on coastal lowlands of the Tuktoyaktuk Peninsula (sites 26–41) were generally <25 m asl.

Most of the lakes were of small to intermediate size. Surface areas ranged from 1.1 (25) to 1262.1 ha (4) (equivalent to 0.011–12.62 km^2), with an average size of 91.4 ha (Table 2). The artificially dammed Fish Lake (4) was eliminated from the PCA because of its relatively large size (1262 ha) and its use as a drinking water reservoir for the Whitehorse area. Thirty-four of the study sites (60%) ranged between 20 and 100 ha whereas 13 (23%) were smaller than 20 ha and only 10 (17%) exceeded 100 ha in size.

The information on maximum depth may not always be adequate and in some cases was only the maximum depth observed during repeated sediment sampling or exploratory soundings. Lake depth ranged from 1.2 (57) to 49 m (6), with a mean of 7.4 m. Most of the deeper lakes (Z_{max} > 5.0 m) were found in the southern Yukon, in particular within the Yukon Plateau area, where lakes were generally confined within valleys. It is not surprising that the forest–tundra and arctic tundra lakes of the Arctic Coastal Plains were relatively shallow, with mean depths of 2.8 and 5.8 m, respectively. This is because these lakes usually occurred in thermokarst depressions. Among those arctic lakes with water depths ≤2.0 m, some may freeze to the bottom during winter, since ice covers ranging in thickness from 1.1 to 2 m have been observed to occur on the Tuktoyaktuk Peninsula (R. Pienitz, unpublished observations).

Lake surface water temperature generally showed a consistent latitudinal gradient of decreasing temperatures from south to north (along with air temperature), with the exception of lakes located in the central part of the Yukon Territory (sites 6–17) where the highest temperatures were recorded. The lowest surface water temperatures were recorded in alpine and arctic tundra lakes, with mean values of 15.5 and 17.1°C, respectively.

Because of their usually small sizes and shallow depths, the average summer surface water temperatures of the lakes surveyed were considerably higher than those reported from previous investigations in the Yukon and on the Tuktoyaktuk Peninsula (see Lindsey et al. 1981; Patalas 1984; Shortreed and Stockner 1986; Anema et al. 1990*a*, 1990*b*). The midsummer temperatures in our lake survey ranged from 12.0 to 23.0°C (Table 2), compared with 8.0–19.8°C (Lindsey et al. 1981), 8.9–14.8°C (Shortreed and Stockner 1986), and 12.5–17.5°C (Patalas 1984). The discrepancy in midsummer surface water temperatures is particularly evident when comparing temperature ranges obtained from lakes on the Tuktoyaktuk Peninsula, with temperatures ranging from 12.0 to 19.0°C, whereas Anema et al. (1990*a*) reported temperatures between 6.8 and 11.7°C in 1985.

Temperature profiles revealed that, in July 1990, complete thermal stratification was established in almost all of our southern Yukon lakes (1–14, 16). Epilimnion depths in these presumably dimictic lakes ranged from 1 to 6 m, usually increasing with lake size and depth (some of the deeper lakes (6, 10) had two thermoclines). Some of the lakes in the forest–tundra near Inuvik (22, 24) were also weakly stratified; however, because of their shallow depths and relatively cool water temperature, a stratification did not develop. Isothermal conditions were observed in our alpine tundra sites (18–21, 58), as well as in some of the smaller and shallower lakes in the boreal forest (15, 17, 56) and forest–tundra (23, 25, 57).

DO concentrations in epilimnetic waters were generally high and ranged from 11.1 to 14.6 $mg \cdot L^{-1}$ (Table 2). DO exhibited an even distribution in most of the lakes, with O_2-saturated waters occurring throughout the water column. In many of these lakes, we recorded a positive heterograde oxygen curve, with higher DO in the metalimnion most likely resulting from photosynthetic activity. Clinograde O_2 curves were observed only in some of the very deep lakes (1, 6), as well as lakes of the Pelly River – Stewart River area (8–11, 13–16) located between Carmacks and Dawson City, where DO levels were greatly reduced in hypolimnetic waters.

Temperature and oxygen profiles could not be measured when sampling from the helicopter, and are therefore not available for the tundra lakes (26–55). In any event, oxygen measurements seem to be redundant in shallow arctic tundra lakes because they are usually completely saturated with oxygen during the open-water season (Hobbie 1984; Welch 1991). Welch (1991) observed that, usually with the beginning of snowmelt, sufficient photosynthesis occurred beneath the lake ice so that O_2 production greatly exceeded O_2 consumption. In addition, thorough mixing generally created well-oxygenated water bodies in tundra regions, where in the mostly shallow lake basins thermoclines were commonly absent owing to the action of strong winds (Fee et al. 1988).

Water transparency (as measured by Secchi disk readings) ranged from 0.7 to 11.5 m (Table 2), which was similar to values of 0.6–13.0 m reported by Lindsey et al. (1981) for Yukon lakes. Mean Secchi depths were highest in boreal forest (4.4 m) and alpine tundra lakes (4.1 m). The low Secchi depths observed in forest–tundra lakes (mean = 1.7 m) were due to the highly coloured (brown) waters of these mostly shallow, low-pH lakes, which were surrounded by muskeg and open spruce woodland (also see below). Secchi readings were only useful in the deeper (Z_{max} > 2.0 m) arctic tundra lakes (about 58%), where Secchi depths ranged from 1.0 to 7.0 m.

The variability in light transmission depends on the amount of suspended inorganic and organic material as well as dissolved organic material (Hobbie 1984) and is reflected by the significant negative correlation between TRANS and such variables as PN, POC, and TPU (Table 3). Water colour results not only from the input of organic compounds from decaying vegetation and the leaching of organic soils in the catchment, but also from natural mineral components such as Fe and Mn (McNeely et al. 1979). Our data suggest that [Fe] appears to have had a particularly strong impact on water colour, given its locally high concentrations (see below) and its significant negative correlation with TRANS (r = –0.75, P < 0.01) (Table 3).

Chemical variables

pH

Measurements of pH indicated that our study sites were

© 1997 NRC Canada

Table 2. Field data collected in 1990 for the 59 lakes.

Lake No.	LAT (°N)	LONG (°W)	ALT (m)	Z_{max} (m)	AREA (ha)	pH	TEMP (°C)	COND ($\mu S \cdot cm^{-1}$)	O_2 ($mg \cdot L^{-1}$)	TRANS (m)	DIST (km)
Boreal forest											
1	60.39	134.57	671	32.0	181.2	8.7	17.5	700	13.7	9.1	na
2	60.40	134.59	671	10.5	56.9	8.8	19.0	1500	14.2	8.5	na
3	60.44	135.02	625	17.5	21.2	8.5	20.0	490	13.7	5.0	na
5	60.42	135.17	1021	12.0	52.5	7.8	17.0	73	13.3	2.3	na
6	61.21	135.39	823	49.0	89.0	8.3	20.0	179	13.3	3.2	na
7	61.42	135.56	634	27.0	163.0	8.3	20.8	230	12.9	10.8	na
8	62.11	136.15	686	5.5	23.7	8.7	23.0	339	14.3	2.7	na
9	62.43	136.41	579	3.0	331.5	8.7	20.7	220	14.4	1.9	na
10	63.01	136.28	655	8.0	88.6	8.6	22.0	242	13.7	4.4	na
11	63.09	136.30	732	5.0	8.6	8.5	23.0	49	11.6	3.3	na
12	63.39	135.54	579	9.2	23.4	8.4	22.0	149	12.2	6.0	na
13	63.59	135.24	701	10.1	10.1	7.8	20.8	45	11.8	3.6	na
14	63.59	135.22	701	16.5	23.1	8.3	21.5	260	13.0	11.5	na
15	63.39	135.51	594	3.0	24.1	7.5	19.4	46	11.8	1.5	na
16	63.45	137.43	610	5.0	80.3	7.6	22.0	42	11.6	2.8	na
17	63.51	138.02	610	4.0	18.9	8.2	19.4	24	12.1	1.7	na
56	67.14	135.26	366	4.0	35.0	7.5	18.0	35	11.4	1.4	na
Forest–tundra											
4	60.37	135.14	1113	7.0	1262.1	8.1	14.0	87	14.6	6.5	na
22	68.11	133.27	76	5.5	8.2	7.9	18.0	153	12.1	4.1	−21.25
23	68.18	133.16	30	2.5	6.8	7.2	18.1	72	12.1	1.8	−7.50
24	68.19	133.22	91	2.8	15.1	6.9	18.0	35	11.1	1.1	−6.25
25	68.24	133.42	122	3.0	1.1	7.8	20.0	140	13.4	**3.0**	0.00
47	68.28	133.38	122	1.5	21.1	8.3	19.5	220	na	**1.5**	7.50
48	68.25	133.35	114	1.5	96.2	8.6	20.5	71	na	1.0	2.50
49	68.23	133.25	84	3.5	5.3	6.6	19.0	41	na	0.7	0.00
50	68.25	133.22	76	2.0	21.7	7.0	20.3	71	na	1.6	5.00
51	68.29	133.22	128	1.5	87.3	8.5	19.5	72	na	1.0	12.50
52	68.34	133.20	145	6.0	19.5	7.0	18.0	116	na	1.8	21.25
57	67.13	135.36	396	1.2	7.2	7.4	18.0	140	13.3	1.0	na
Arctic tundra											
26	69.28	132.49	18	2.0	85.1	8.1	17.0	100	na	**2.0**	125.00
27	69.32	132.47	15	2.0	94.6	8.0	17.0	343	na	1.0	132.50
28	69.33	132.45	15	2.0	170.3	7.8	17.0	145	na	1.2	135.00
29	69.33	132.25	15	2.0	547.4	8.1	17.0	152	na	**2.0**	135.00
30	69.35	132.04	21	4.0	195.8	8.0	16.0	179	na	3.2	137.50
31	69.32	132.04	21	3.0	57.6	8.1	16.0	165	na	2.8	133.75
32	69.28	132.12	21	9.0	73.8	8.0	12.0	198	na	6.0	126.25
33	69.29	132.19	15	2.0	5.4	7.3	19.0	87	na	1.0	126.25
34	69.25	132.40	24	2.5	116.2	7.9	17.0	159	na	**2.5**	118.75
35	69.19	132.59	24	7.0	86.3	8.2	17.0	128	na	2.7	107.50
36	69.10	133.16	21	4.0	103.2	8.4	17.0	105	na	3.2	87.50
37	69.08	133.17	21	6.5	69.0	8.1	17.0	98	na	4.0	83.75
38	69.07	133.11	21	3.0	49.7	8.2	18.0	139	na	**3.0**	82.50
39	69.12	133.02	24	12.0	104.9	8.2	15.5	141	na	6.0	91.25
40	69.13	133.00	24	3.0	81.9	8.2	17.5	104	na	2.5	92.50
41	69.20	132.44	24	7.0	85.5	8.1	16.0	167	na	2.5	108.75
42	69.03	133.27	46	15.0	84.8	7.8	16.0	150	na	7.0	72.50
43	68.59	133.28	30	14.0	36.0	7.5	17.0	105	na	7.0	66.25
44	68.50	133.33	30	4.0	61.2	8.2	18.5	129	na	2.8	47.50
45	68.46	133.39	76	2.0	65.4	6.9	19.0	65	na	1.5	41.25
46	68.29	133.39	122	2.5	63.1	7.1	19.0	81	na	**2.5**	10.00
53	68.36	133.15	152	3.5	25.7	7.2	18.2	48	na	2.3	25.00

© 1997 NRC Canada

Table 2 (*concluded*).

Lake No.	LAT (°N)	LONG (°W)	ALT (m)	Z_{max} (m)	AREA (ha)	pH	TEMP (°C)	COND ($\mu S \cdot cm^{-1}$)	O_2 ($mg \cdot L^{-1}$)	TRANS (m)	DIST (km)
54	68.38	133.17	91	10.0	40.0	7.3	18.0	85	na	1.9	30.00
55	68.42	133.15	30	18.5	87.9	7.7	18.0	70	na	3.5	37.50
Alpine tundra											
18	64.35	138.18	1173	7.5	18.1	8.6	17.0	113	12.7	4.6	na
19	64.39	138.23	1128	3.8	20.4	8.7	16.0	111	13.2	3.2	na
20	64.44	138.22	1097	1.9	13.7	9.3	14.3	39	12.8	**1.9**	na
21	64.51	138.21	1006	3.8	144.0	7.5	17.5	32	11.7	3.7	na
58	67.06	136.00	549	5.5	19.3	5.9	16.2	77	12.9	1.9	na
59	64.29	138.17	1387	15.5	4.2	7.9	13.5	65	na	9.5	na
Minimum	60.37	132.04	15	1.2	1.1	5.9	12.0	24	11.1	0.7	–21.25
Maximum	69.35	138.23	1387	49.0	1262.1	9.3	23.0	1500	14.6	11.5	137.50
Median			122	4.0	56.9	8.1	18.0	111	12.9	2.7	66.25
Mean			355.9	7.4	91.4	7.9	18.2	159.7	12.8	3.4	63.75

Note: na, not available; TRANS, Secchi disk transparency (bold numbers = readings where TRANS = Z_{max}); DIST, shortest distance from northern treeline (defined as 80% tree cover).

slightly acidic to alkaline, ranging from a pH of 5.9 (58) to 9.3 (20) (Table 2). The lowest pH values were recorded in the forest–tundra and tundra near Inuvik, where most of the surface waters were circumneutral to slightly acidic (e.g., sites 22–25, 45, 46, 49, 50, 52–54). The highest pH values were recorded in the southern Yukon, particularly in the Whitehorse area. PCA ordination and the Pearson correlation matrix showed a strong positive correlation between pH and DIC ($r = 0.71$, $P < 0.01$), as opposed to a strong negative correlation between pH and Fe ($r = -0.54$, $P < 0.01$) (Table 3; Fig. 2).

Conductivity and major ions

Patterns in the conductivity data follow those observed in most of the major ions. For example, the major ions Na, Cl, K, Ca, and SO_4, as well as DIC, are highly correlated with conductivity ($P < 0.01$) (Table 3; Fig. 2). A PCA of ion data with COND demonstrated that all of these variables are significantly negatively correlated with latitude and related climatic variables.

Ca levels were usually high and ranged from 3.5 (17) to 50.3 $mg \cdot L^{-1}$ (7), with an average of 19.6 $mg \cdot L^{-1}$ (Table 4). About 50% of the lakes had concentrations exceeding this average, indicating that most Yukon lakes are alkaline, Ca-rich waters. Not surprisingly, [Ca] is significantly correlated with [DIC] and pH (Table 3, $P < 0.05$).

High SO_4 concentrations are generally due to weathering of sedimentary bedrock and are particularly evident in lakes of the Whitehorse area (e.g., sites 1–3), where rocks composed of limestone–dolomite, sandstone, siltstone, and conglomerate prevail (Oswald and Senyk 1977). Slightly elevated concentrations of SO_4 were also observed in the vicinity of Inuvik.

Except for lakes 1 and 2 in the Whitehorse area, Cl levels were generally low in boreal forest (mean = 5.0 $mg \cdot L^{-1}$), forest–tundra (mean = 2.7 $mg \cdot L^{-1}$), and alpine tundra (mean = 0.4 $mg \cdot L^{-1}$) lakes. In contrast, [Cl] was appreciably higher in most of the arctic tundra lakes with a mean of 12.2 $mg \cdot L^{-1}$. Within this subset of lakes, [Cl] clearly varied as a function of distance from the Arctic Ocean (Fig. 3), with highest concentrations occurring in lakes located on coastal plains in the vicinity of Tuktoyaktuk (in particular, lakes 26–41). This trend is also reflected in the significant correlation between [Cl] and TUNDRA ($r = 0.61$, $P < 0.01$) (Table 3).

A similarly strong relationship between [Cl] and distance from the coast has been observed in lakes of the Kenai Peninsula (Eilers et al. 1993) and the North Slope of Alaska (Livingstone 1966; Kalff 1968; Kling et al. 1992). Sea salt in precipitation is likely to be the main source of Cl in lakes of these maritime arctic regions. However, it is possible that edaphic factors, related to the emergence of the coastal lowlands from postglacial marine waters may also play an important role in the distribution of Cl^- (see Welch and Legault 1986).

Na concentrations were highly correlated with those of Cl (Fig. 4) and decreased with distance from the coast in a pattern similar to that observed for Cl.

Nutrients and carbon

Concentrations of SiO_2 were low to moderate and ranged from 0.1 to 12.5 $mg \cdot L^{-1}$, with a mean of 2.1 $mg \cdot L^{-1}$ for all lakes. Average concentrations calculated for boreal forest, forest–tundra, and arctic tundra lakes showed a latitudinal gradient, with values decreasing with increasing latitude from 4.7 to 1.5 and 0.7 $mg \cdot L^{-1}$, respectively. This trend was corroborated by the significant negative correlation between SiO_2 and LAT ($r = -0.62$, $P < 0.01$) (Table 3; Fig. 2). SiO_2 is most easily leached from sedimentary and volcanic rocks (McNeely et al. 1979; Prentki et al. 1980) that predominate in the southern Yukon, and where the highest SiO_2 concentrations were recorded in lakes 3–10 (Table 4).

The concentrations of DIC showed considerable variation among lakes and ranged from 0.3 (58) to 134.2 $mg \cdot L^{-1}$ (2)

© 1997 NRC Canada

Table 3. Pearson correlation matrix for environmental variables (following transformations) measured in the 59 lakes included in the PCA.

	LAT	LONG	ALT	DEPTH	AREA	TEMP	TRANS	COND	pH	CHLAU	CHLAC	TPU	TPF	SRP	NO_2	NO_3	NH_3
LONG	−0.73**																
ALT	−0.88	0.88**															
DEPTH	−0.52	0.22	0.35														
AREA	−0.04	−0.20	−0.20	0.13													
TEMP	−0.35	0.20	0.32	0.00	−0.21												
TRANS	−0.44	0.22	0.30	0.74**	0.13	−0.08											
COND	−0.21	−0.21	−0.07	0.34	0.26	0.05	0.46										
pH	−0.39	0.26	0.21	0.20	0.28	0.08	0.33	0.46									
CHLAU	0.10	0.05	0.05	−0.36	−0.10	0.22	−0.36	−0.37	−0.02								
CHLAC	0.14	0.05	0.04	−0.40	−0.16	0.18	−0.37	−0.28	0.05	0.87**							
TPU	0.17	−0.04	0.00	−0.54**	−0.19	0.23	−0.53**	−0.36	−0.16	0.62**	0.64**						
TPF	0.15	−0.08	−0.02	−0.41	−0.35	0.16	−0.40	−0.37	−0.35	0.24	0.24	0.77**					
SRP	−0.21	0.06	0.18	0.01	−0.16	0.16	0.04	0.08	0.01	−0.02	−0.02	0.37	0.58**				
NO_2	0.12	−0.10	−0.13	0.14	−0.07	−0.09	0.17	−0.07	−0.14	−0.06	−0.02	−0.06	−0.02	0.04			
NO_3	−0.19	−0.04	0.08	0.31	0.16	−0.06	0.31	0.32	0.12	−0.19	−0.12	−0.00	0.10	0.13	0.07		
NH_3	0.30	−0.41	−0.37	−0.36	0.03	−0.02	−0.20	0.19	−0.06	−0.11	−0.07	0.15	0.19	0.08	−0.01	0.01	
TKN	−0.16	0.13	0.11	−0.39	−0.20	0.58**	−0.36	−0.01	0.13	0.22	0.20	0.44	0.35	0.30	−0.11	−0.09	0.36
TN	−0.16	0.15	0.14	−0.41	−0.13	0.56**	−0.39	−0.06	0.15	0.39	0.39	0.58**	0.37	0.27	−0.09	−0.02	0.31
PN	0.01	0.17	0.10	−0.50*	−0.06	0.41	−0.53**	−0.32	0.01	0.69**	0.68**	0.74**	0.36	0.05	−0.08	−0.16	0.10
POC	−0.06	0.20	0.15	−0.45	−0.06	0.45	−0.50*	−0.28	0.02	0.67**	0.64**	0.74**	0.36	0.10	−0.09	−0.14	0.11
DOC	−0.19	0.12	0.19	−0.26	−0.25	0.61**	−0.28	−0.08	−0.02	0.17	0.08	0.44	0.42	0.33	−0.09	−0.05	0.31
DIC	−0.33	−0.08	0.00	0.44	0.39	0.04	0.54**	0.86**	0.71**	−0.36	−0.33	−0.42	−0.43	0.09	−0.06	0.27	0.09
SiO_2	−0.62**	0.30	0.51*	0.55**	0.09	0.24	0.42	0.32	0.14	−0.16	−0.20	−0.20	−0.22	0.09	−0.06	0.09	−0.28
Cl	0.43	−0.68**	−0.71**	−0.11	0.33	−0.27	0.00	0.51*	0.15	−0.30	−0.21	−0.12	−0.04	0.01	0.02	0.36	0.49
SO_4	−0.37	0.06	0.32	0.27	−0.10	0.13	0.32	0.68**	0.07	−0.26	−0.17	−0.09	−0.02	0.29	−0.18	0.30	0.12
Na	−0.02	−0.41	−0.28	0.15	0.33	−0.02	0.24	0.86**	0.35	−0.40	−0.33	−0.21	−0.17	0.14	−0.08	0.31	0.44
K	−0.32	−0.13	0.05	0.22	0.22	0.27	0.32	0.78**	0.37	−0.20	−0.12	−0.05	0.01	0.34	−0.09	0.41	0.29
Ca	−0.18	−0.06	−0.02	0.37	0.25	0.05	0.42	0.69**	0.51*	−0.30	−0.27	−0.46	−0.53*	−0.10	−0.13	0.00	−0.03
Fe	0.53**	−0.22	−0.30	−0.66	−0.29	−0.07	−0.75**	−0.59**	−0.54**	0.38	0.36	0.70**	0.68**	0.11	−0.08	−0.19	0.26
Mn	0.36	−0.04	−0.10	−0.32	−0.12	−0.02	−0.36	−0.23	−0.46	0.20	0.25	0.45	0.34	0.09	0.10	−0.07	0.14
FOREST	−0.80**	0.49*	0.63**	0.46	0.01	0.62**	0.31	0.20	0.33	0.07	0.02	−0.07	−0.10	0.18	−0.12	0.15	−0.20
WOOD	0.18	−0.20	−0.03	−0.35	−0.30	0.09	−0.28	−0.15	−0.26	0.27	0.32	0.52*	0.50*	0.23	0.00	−0.04	0.01
TUNDRA	0.70**	−0.69**	−0.83**	−0.14	0.37	−0.41	−0.12	0.08	−0.11	−0.17	−0.20	−0.28	−0.25	−0.32	0.14	−0.06	0.30
ALPINE	−0.17	0.64**	0.43	0.00	−0.21	−0.37	0.09	−0.24	0.03	−0.18	−0.12	−0.13	−0.10	−0.06	−0.05	−0.08	−0.20

Note: Significant correlations based on Bonferroni-adjusted probabilities: *, $P \leq 0.05$; **, $P \leq 0.01$.

(Table 4). This corresponds to an alkalinity range of approximately 0–2.5 mequiv. $\cdot L^{-1}$, as calculated from [DIC] and pH using the carbonate species relations in Stumm and Morgan (1981). The highest DIC concentrations were observed in boreal forest lakes near Whitehorse, where values often exceeded 30 mg $\cdot L^{-1}$. They were generally lowest in forest–tundra and alpine tundra lakes.

The discrepancy between DOC concentrations in lakes with forested and unforested watersheds was evident (Table 4; Fig. 5). The lowest average [DOC] was observed in alpine (7.9 mg $\cdot L^{-1}$) and arctic tundra (9.4 mg $\cdot L^{-1}$) sites whereas concentrations were considerably higher in boreal forest and forest–tundra lakes (15.9 mg $\cdot L^{-1}$). This strong relationship between [DOC] and catchment vegetation is represented by the significant negative correlation ($r = -0.49$, $P < 0.05$, Table 3) and the PCA ordination plot (Fig. 2). Despite the relatively high concentrations of DOC in some of the study lakes, none was strongly acidic (minimum measured pH = 5.9, lake 58). Most of the high-DOC lakes received organic inputs from bog vegetation and *Sphagnum* mosses in their watersheds. However, the production of organic acids from these sources seemed to be insufficient to overcome the alkalinity produced from weathering processes in the watershed and in-lake processes.

Of the four nitrogen species analysed, TKN (dissolved organic nitrogen, measured on the filtered fraction) was the most variable, with highest mean concentrations in boreal forest (563 $\mu g \cdot L^{-1}$) and forest–tundra lakes (557 $\mu g \cdot L^{-1}$). Values for arctic and alpine tundra lakes were considerably lower, with 350 and 263 $\mu g \cdot L^{-1}$, respectively. Values ranged from a low of 72 $\mu g \cdot L^{-1}$ (59) to 1293 $\mu g \cdot L^{-1}$ (8) and averaged 439 $\mu g \cdot L^{-1}$ for all lakes (Table 4). As expected, TKN was highly correlated with TN ($r = 0.96$, $P < 0.01$) (Table 3), which was calculated as the sum of PN, TKN, NO_3, and NO_2.

[TN] ranged from a low of 123 $\mu g \cdot L^{-1}$ (59) to 1585 $\mu g \cdot L^{-1}$ (9), this range being typical of oligotrophic waters according to Wetzel (1983). Average summer surface water concentrations were almost equally high in forest–tundra and boreal forest lakes (730 and 722 $\mu g \cdot L^{-1}$, respectively) whereas they were substantially lower in arctic (435 $\mu g \cdot L^{-1}$) and alpine tundra lakes (343 $\mu g \cdot L^{-1}$).

The concentrations for the three species of inorganic nitrogen (NO_3, NO_2, and NH_3) were below or near the analytical detection limit in many lakes. NO_3 values ranged from 9 to 208 $\mu g \cdot L^{-1}$, NO_2 from 0.2 to 29 $\mu g \cdot L$, and NH_3 from 5 to 51 $\mu g \cdot L^{-1}$. The highest NO_3 concentration was recorded in lake 1 (208 $\mu g \cdot L^{-1}$) (Table 4).

Among the phosphorus variables SRP, TPU, and TPF, [TPU] and [TPF] were highly correlated ($r = 0.77$, $P < 0.01$)

© 1997 NRC Canada

Table 3 (*continued*).

TKN	TN	PN	POC	DOC	DIC	SiO_2	Cl	SO_4	Na	K	Ca	Fe	Mn	FOREST	WOOD	TUNDRA
0.96**																
0.58**	0.76**															
0.63**	0.79**	0.99**														
0.89**	0.84**	0.46	0.52*													
0.03	−0.03	−0.34	−0.30	−0.04												
0.02	−0.01	−0.20	−0.14	0.08	0.38											
−0.03	−0.07	−0.22	−0.22	−0.11	0.43	−0.41										
0.13	0.09	−0.15	−0.11	0.17	0.41	0.42	0.09									
0.10	0.03	−0.26	−0.23	0.01	0.75**	0.07	0.72**	0.55**								
0.24	0.24	0.02	0.05	0.15	0.66**	0.13	0.50*	0.62**	0.80**							
−0.03	−0.11	−0.36	−0.34	−0.04	0.77**	0.50*	0.21	0.34	0.45	0.28						
0.18	0.23	0.43	0.41	0.22	−0.72**	−0.48	−0.02	−0.30	−0.32	−0.38	−0.60**					
0.10	0.17	0.35	0.33	0.11	−0.46	−0.12	−0.18	−0.05	−0.18	−0.22	−0.33	0.53*				
0.34	0.34	0.20	0.27	0.37	0.31	0.46	−0.26	0.24	0.05	0.42	0.20	−0.46	−0.36			
0.17	0.21	0.15	0.12	0.21	−0.31	0.06	−0.16	0.18	−0.14	−0.12	−0.16	0.37	0.28	−0.32		
−0.30	−0.34	−0.24	−0.28	−0.37	0.09	−0.45	0.61**	−0.37	0.26	−0.05	0.02	0.08	−0.02	−0.53*	−0.42	
−0.24	−0.24	−0.10	−0.11	−0.49*	−0.20	−0.03	−0.38	0.00	−0.33	−0.38	−0.12	0.05	0.19	−0.21	−0.17	−0.28

(Table 3). [TPU] ranged from 3.0 $\mu g \cdot L$ (32) to 55.1 $\mu g \cdot L^{-1}$ (51) and averaged 15.5 $\mu g \cdot L^{-1}$ for all lakes (Table 4). Interestingly, the average TPU value obtained for lakes in the forest–tundra near Inuvik (29.5 $\mu g \cdot L^{-1}$) was more than double that recorded for boreal forest lakes (14.2 $\mu g \cdot L^{-1}$). North of and above the treeline, average TPU concentrations were almost equally low in arctic (11.6 $\mu g \cdot L^{-1}$) and alpine tundra lakes (11.0 $\mu g \cdot L^{-1}$).

On the basis of the general relationship between lake productivity and average concentrations of epilimnetic TP established by Vollenweider (1968, 1976), we conclude that most of our study lakes (52%) fall within the mesoeutrophic and oligomesotrophic range (24%). These results are unexpected, considering that TP concentrations in arctic lakes are usually low (Wetzel 1983; Hobbie 1984). Locally high TP concentrations within our study area may be related to the diverse geological and edaphic conditions (i.e., soil development) in the Yukon, where sedimentary rock deposits (e.g., siltstones, sandstones, and shales of Mesozoic age; limestones and dolomites of Devonian and Ordovician age) are widespread.

Concentrations of SRP were generally low ($\leq$3.3 $\mu g \cdot L^{-1}$), with an average concentration of 1.5 $\mu g \cdot L^{-1}$ for all lakes. Values were especially low in arctic tundra lakes (mean = 0.9 $\mu g \cdot L^{-1}$), where concentrations frequently decreased to less than detectable levels (0.5 $\mu g \cdot L^{-1}$). [SRP] was also low in Barrow (Alaska) lakes, where concentrations ranged between 0.5 and 3.0 $\mu g \cdot L^{-1}$ (Prentki et al. 1980; Hobbie 1984).

TN:TP ratios ranged from 13:1 to 103:1 (mean = 45:1), which was comparable to ratios reported by Shortreed and Stockner (1986) for 19 lakes in the Yukon. The highest TN:TP ratios were generally observed in boreal forest lakes of the southern Yukon (mean = 60:1), with particularly high ratios recorded in lakes of the Pelly Crossing area in the central Yukon (sites 8–10) (Table 4). TN:TP ratios were considerably lower in forest–tundra, arctic tundra, and alpine tundra sites, with averages ranging from 33:1 to 42:1.

Iron and manganese

The average concentrations of Fe and Mn were 210.7 and 23.5 $\mu g \cdot L^{-1}$, respectively (Table 4). Average [Mn] ranged from 12.1 $\mu g \cdot L^{-1}$ in the boreal forest to 42.7 $\mu g \cdot L^{-1}$ in the alpine tundra lakes. In contrast, [Fe] averaged 564.9 $\mu g \cdot L^{-1}$ in forest–tundra lakes and reached highest concentrations of 1660 (49) and 1280 $\mu g \cdot L^{-1}$ (24) in lakes near Inuvik. The Fe concentrations in this area were particularly high in strongly coloured lakes receiving drainage from highly organic soils of surrounding bogs and peatlands. As the humic and fulvic acids of such organic soils form strong soluble complexes with multivalent cations such as Fe and Mn (Schnitzer and Desjardins 1969), the high concentrations of these elements may result from increased mobilization of organometallic complexes in peaty soils. According to Hobbie (1973), high inputs of Fe are due mainly to chelation by dissolved organic matter. The strong relationship between water colour and [Fe] is reflected

© 1997 NRC Canada

Table 4. Water chemistry data (1990) for the 59 lakes.

Lake No.	TPU ($\mu g \cdot L^{-1}$)	TPF ($\mu g \cdot L^{-1}$)	SRP ($\mu g \cdot L^{-1}$)	NO_2 ($\mu g \cdot L^{-1}$)	NO_3 ($\mu g \cdot L^{-1}$)	NH_3 ($\mu g \cdot L^{-1}$)	DOC ($mg \cdot L^{-1}$)	DIC ($mg \cdot L^{-1}$)	TKN ($\mu g \cdot L^{-1}$)	TN ($\mu g \cdot L^{-1}$)	SiO_2 ($mg \cdot L^{-1}$)	SO_4 ($mg \cdot L^{-1}$)	Ca ($mg \cdot L^{-1}$)
Boreal forest													
1	14.1	11.0	2.2	0.3	208.0	12.0	10.6	88.2	309	559	2.81	300.2	21.50
2	15.8	14.1	7.5	1.0	15.0	31.0	16.3	134.2	654	734	0.78	1242.0	15.60
3	6.5	4.5	0.5	1.0	14.0	7.0	7.8	52.9	407	514	12.47	82.1	26.70
5	8.7	4.9	ADL	1.4	ADL	ADL	8.6	12.4	260	323	7.86	3.9	16.60
6	5.1	4.3	3.3	ADL	ADL	ADL	10.1	33.1	271	321	9.26	24.5	44.20
7	4.9	3.5	0.6	ADL	ADL	ADL	8.4	40.8	208	259	9.19	29.0	50.30
8	14.6	0.2	1.8	0.2	ADL	6.0	35.1	53.5	1293	1403	4.98	90.5	37.60
9	15.4	0.2	1.3	0.6	ADL	27.0	26.9	30.8	1178	1585	7.26	23.8	37.70
10	12.0	8.1	2.0	ADL	ADL	ADL	14.2	35.1	743	885	6.54	28.9	30.00
11	12.3	9.0	1.6	0.5	ADL	5.0	17.1	3.8	572	667	0.19	2.0	7.78
12	8.7	5.5	1.1	ADL	ADL	ADL	13.5	22.5	352	418	3.08	0.5	31.00
13	9.8	5.6	1.2	1.5	ADL	ADL	16.2	4.6	437	521	0.23	2.8	7.27
14	4.3	2.9	2.4	0.9	ADL	6.0	15.9	35.1	350	409	8.84	57.1	45.40
15	28.8	13.9	2.1	2.9	ADL	5.0	24.2	4.2	699	1036	0.16	4.2	8.55
16	21.6	11.4	1.3	ADL	ADL	ADL	15.3	4.9	615	769	1.00	0.5	6.84
17	25.0	13.0	1.3	ADL	ADL	5.0	18.1	2.2	904	1258	0.17	0.5	3.52
56	35.4	14.5	1.6	1.0	ADL	14.0	12.3	2.4	409	620	1.02	7.6	5.48
Forest–tundra													
4	3.7	2.9	1.4	0.9	ADL	ADL	3.8	14.4	121	158	7.84	8.8	19.00
22	10.3	7.9	1.6	0.9	ADL	ADL	17.8	14.4	532	591	3.16	38.9	39.20
23	17.5	11.3	2.3	1.6	ADL	ADL	10.6	4.6	637	717	0.94	13.2	13.90
24	44.9	34.1	10.1	4.3	27.0	11.0	22.6	4.0	700	803	2.37	5.2	6.50
25	16.1	12.1	2.0	0.8	ADL	22.0	13.1	11.5	631	710	0.57	32.6	20.20
47	9.3	6.0	ADL	ADL	15.0	ADL	9.1	18.9	334	401	0.94	42.3	33.90
48	48.8	12.7	3.2	1.4	15.0	ADL	11.6	5.5	518	1058	0.39	8.1	9.23
49	43.9	21.2	2.0	0.6	16.0	ADL	21.5	2.0	612	781	3.31	9.1	6.73
50	28.6	24.3	0.6	2.9	16.0	37.0	29.9	3.9	823	922	0.75	9.3	9.17
51	55.1	13.6	0.6	1.9	16.0	26.0	17.2	7.4	688	1114	1.73	3.8	10.20
52	9.7	6.6	0.6	1.5	14.0	16.0	11.3	1.8	316	388	1.54	49.4	12.60
57	40.5	16.8	1.4	ADL	ADL	6.0	9.8	2.1	335	545	0.51	31.9	9.49
Arctic tundra													
26	15.3	7.1	1.5	1.1	ADL	19.0	6.6	6.3	305	380	0.14	1.9	9.14
27	19.8	8.0	2.2	ADL	15.0	36.0	12.3	27.7	535	630	0.32	17.9	43.80
28	16.7	8.1	0.7	0.4	ADL	ADL	6.7	12.9	281	403	0.29	6.0	19.30
29	9.2	4.8	ADL	ADL	ADL	ADL	5.8	17.5	239	309	0.10	1.1	20.80
30	12.6	6.0	ADL	ADL	ADL	38.0	18.2	19.6	567	649	0.09	24.7	26.90
31	6.6	3.7	ADL	ADL	10.0	18.0	8.0	19.2	343	401	0.17	10.5	24.70
32	3.0	2.9	ADL	0.9	15.0	8.0	4.8	27.4	172	220	1.52	9.2	36.20
33	13.8	11.4	1.9	1.9	11.0	51.0	19.0	6.8	862	955	0.40	4.7	11.50
34	13.7	6.3	ADL	ADL	ADL	20.0	10.7	17.9	391	473	0.57	3.0	24.00
35	11.3	6.6	ADL	ADL	ADL	22.0	8.5	17.5	282	364	0.42	2.8	21.80
36	20.8	6.2	0.7	ADL	ADL	19.0	9.6	14.1	459	541	0.43	1.4	18.30
37	10.5	5.6	ADL	0.6	ADL	17.0	8.9	12.7	309	404	0.51	1.4	16.40
38	13.3	5.3	ADL	ADL	ADL	15.0	9.6	20.8	384	506	0.57	2.1	24.20
39	5.0	3.7	ADL	ADL	ADL	ADL	6.1	21.4	211	273	1.37	4.7	29.60
40	13.1	10.3	ADL	ADL	ADL	6.0	10.1	13.3	497	587	0.64	0.8	18.90
41	7.5	4.2	ADL	ADL	ADL	9.0	7.9	24.0	247	305	0.61	9.2	32.20
42	3.5	2.7	2.2	7.2	51.0	10.0	4.6	15.7	139	229	1.54	1.9	20.80
43	5.0	3.2	0.5	29.0	23.0	9.0	5.7	10.3	197	305	0.81	1.6	14.00
44	12.3	6.6	1.1	3.7	30.0	8.0	8.1	14.2	336	451	0.73	2.1	19.80
45	17.8	12.6	ADL	0.2	20.0	14.0	11.8	3.4	397	497	1.92	12.9	7.78
46	16.2	10.9	0.6	ADL	14.0	19.0	11.4	4.9	439	541	1.18	14.6	9.74
53	11.4	7.8	1.4	1.4	ADL	21.0	10.8	3.7	263	341	0.96	6.1	5.89

© 1997 NRC Canada

Table 4 (*continued*).

Na ($mg \cdot L^{-1}$)	K ($mg \cdot L^{-1}$)	Cl ($mg \cdot L^{-1}$)	CHLAU ($\mu g \cdot L^{-1}$)	CHLAC ($\mu g \cdot L^{-1}$)	POC ($\mu g \cdot L^{-1}$)	PN ($\mu g \cdot L^{-1}$)	Fe ($\mu g \cdot L^{-1}$)	Mn ($\mu g \cdot L^{-1}$)	TN:TP	POC:PN
46.30	10.10	63.6	0.3	ADL	321	42	8.4	5.0	40	8
187.00	29.90	24.5	0.1	ADL	493	64	11.4	12.0	46	8
16.30	4.30	1.5	1.5	ADL	704	92	10.8	7.0	79	8
1.95	0.56	0.6	2.3	1.3	369	52	76.0	4.0	37	7
4.60	1.21	0.9	0.3	ADL	354	40	15.8	12.0	63	9
7.56	1.85	0.9	0.2	ADL	279	41	5.9	7.0	53	7
20.70	1.93	1.4	1.5	0.7	848	100	19.4	16.0	96	8
4.32	2.84	1.3	1.9	1.3	3280	396	17.4	21.0	103	8
4.36	2.94	1.5	0.9	0.7	828	132	6.3	17.0	74	6
0.73	1.57	4.2	0.9	0.7	592	84	50.2	ADL	54	7
1.32	1.45	1.0	2.8	1.0	412	56	26.5	11.0	48	7
0.39	0.46	0.5	1.7	0.7	495	72	36.5	4.0	53	7
3.69	1.60	0.4	2.7	ADL	398	48	10.0	7.0	95	8
0.53	1.10	0.9	4.9	1.1	2170	324	165.0	18.0	36	7
0.94	1.57	0.5	2.5	0.8	1060	144	124.0	21.0	36	7
0.75	1.26	0.7	4.9	2.3	2320	344	134.0	19.0	50	7
0.77	0.84	0.7	10.5	8.0	1380	200	612.0	23.0	18	7
2.80	0.77	0.3	1.7	ADL	158	26	8.6	5.0	43	6
2.26	1.04	6.1	1.8	1.3	300	48	52.7	11.0	57	6
1.60	0.65	3.7	2.0	1.4	472	68	287.0	11.0	41	7
1.32	0.80	2.0	1.1	ADL	536	72	1280.0	33.0	18	7
8.28	2.07	3.2	2.6	2.1	464	68	136.0	20.0	44	7
5.02	1.64	2.7	0.7	0.4	272	52	26.7	8.0	43	5
1.60	1.38	1.6	20.4	18.1	3480	524	272.0	39.0	22	7
1.38	0.45	1.4	1.8	0.7	1180	152	1660.0	61.0	18	8
4.93	1.10	5.2	0.6	0.2	572	80	774.0	50.0	32	7
4.21	1.22	2.0	4.9	4.0	2940	408	771.0	43.0	20	7
7.23	1.14	1.7	0.5	0.1	304	56	348.0	72.0	40	5
0.85	1.05	0.5	7.9	6.7	1340	200	607.0	44.0	13	7
8.16	1.11	15.5	1.5	0.7	416	64	170.0	14.0	25	7
33.40	3.23	75.2	1.4	0.6	544	80	754.0	16.0	32	7
9.42	1.79	18.8	1.1	0.5	780	112	376.0	13.0	24	7
10.30	0.90	18.7	0.8	0.2	444	60	65.5	11.0	34	7
10.40	1.22	21.5	1.8	ADL	504	72	80.6	11.0	52	7
8.97	1.43	16.3	0.8	0.3	364	48	44.6	7.0	61	8
7.63	1.36	13.2	0.2	ADL	204	32	15.6	6.0	73	6
6.05	1.14	10.0	0.9	0.3	572	80	297.0	13.0	69	7
9.72	1.43	17.3	1.8	0.5	492	72	153.0	46.0	35	7
6.10	1.63	10.1	1.9	1.3	440	72	143.0	11.0	32	6
4.71	1.15	7.2	1.8	1.1	480	72	92.5	18.0	26	7
4.58	1.02	6.8	1.0	0.3	548	84	58.0	13.0	38	7
7.27	1.31	8.9	1.9	1.0	616	112	38.9	16.0	38	6
5.04	1.16	7.4	1.4	0.6	320	52	24.2	7.0	55	6
4.86	1.15	8.2	2.6	1.4	496	80	165.0	18.0	45	6
7.83	1.96	12.2	1.0	0.6	304	48	51.7	9.0	41	6
3.36	0.89	5.2	1.1	0.6	228	32	21.1	160.0	65	7
3.08	0.98	5.6	1.0	0.6	352	56	19.1	14.0	61	6
3.08	1.10	3.7	1.2	0.7	456	81	72.0	19.0	37	6
2.22	0.90	1.9	1.4	0.9	508	80	348.0	23.0	28	6
2.39	1.39	1.9	1.1	0.6	588	88	235.0	33.0	33	7
3.06	0.77	1.6	1.3	0.6	468	68	110.0	14.0	30	7

© 1997 NRC Canada

Table 4 (*concluded*).

Lake No.	TPU ($\mu g \cdot L^{-1}$)	TPF ($\mu g \cdot L^{-1}$)	SRP ($\mu g \cdot L^{-1}$)	NO_2 ($\mu g \cdot L^{-1}$)	NO_3 ($\mu g \cdot L^{-1}$)	NH_3 ($\mu g \cdot L^{-1}$)	DOC ($mg \cdot L^{-1}$)	DIC ($mg \cdot L^{-1}$)	TKN ($\mu g \cdot L^{-1}$)	TN ($\mu g \cdot L^{-1}$)	SiO_2 ($mg \cdot L^{-1}$)	SO_4 ($mg \cdot L^{-1}$)	Ca ($mg \cdot L^{-1}$)
54	11.7	7.5	1.4	0.8	ADL	ADL	13.0	5.1	301	364	1.26	19.0	11.90
55	9.0	7.2	0.5	ADL	ADL	ADL	8.5	7.4	237	319	0.62	3.2	10.40
Alpine tundra													
18	11.4	6.2	1.0	0.8	ADL	ADL	6.5	18.1	239	302	2.03	11.5	23.00
19	10.0	6.9	1.4	0.8	ADL	ADL	10.2	20.5	348	415	1.95	9.0	31.60
20	23.1	14.1	2.4	2.0	ADL	11.0	12.3	5.7	498	606	0.37	1.9	9.66
21	7.8	4.9	1.2	0.6	ADL	ADL	11.4	3.8	327	422	0.30	4.1	5.82
58	9.8	4.5	ADL	ADL	ADL	ADL	3.9	0.3	93	191	1.92	39.0	8.29
59	4.0	2.8	0.5	0.9	ADL	ADL	3.1	5.5	72	123	1.74	15.2	12.80
Minimum	3.0	0.2	0.5	0.2	9.0	5.0	3.1	0.3	72	123	0.09	0.5	3.52
Maximum	55.1	34.1	10.1	29.0	208.0	51.0	35.1	134.2	1293	1585	12.47	1242.0	50.30
Median	12.3	6.6	1.2	0.6	10.0	6.0	10.7	12.9	352	497	0.94	8.8	18.30
Mean	15.5	8.4	1.5	1.4	16.0	12.2	12.3	17.8	438.6	558.9	2.12	40.6	19.58

Note: ADL, at detection limit; TPU, total phosphorus (unfiltered); TPF, total phosphorus (filtered); SRP, soluble reactive phosphorus; CHLAU, chlorophyll *a* (uncorrected); CHLAC, chlorophyll *a* (corrected).

Fig. 3. Plot of Cl^- concentrations versus distance from the Arctic Ocean in a subset of 31 lakes from the Inuvik area and the Tuktoyaktuk Peninsula.

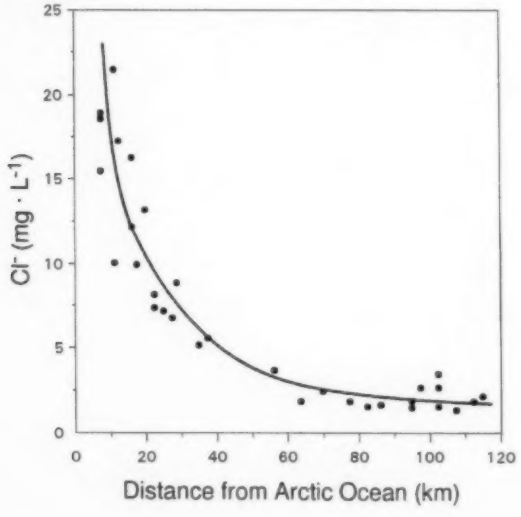
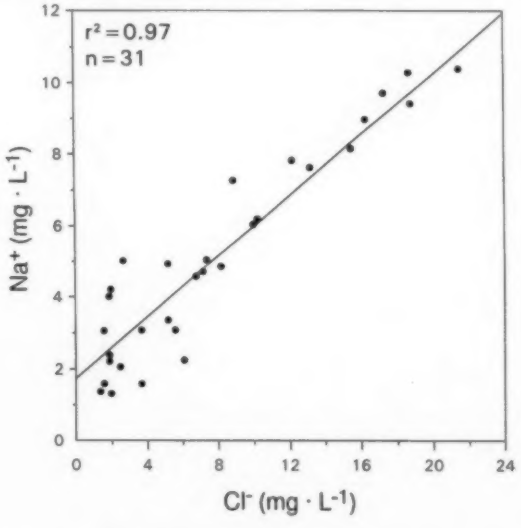


Fig. 4. Plot of Na^+ versus Cl^- concentrations in a subset of 31 lakes from the Inuvik area and the Tuktoyaktuk Peninsula. The line shows a relationship that is expected if the concentrations of both ions were derived entirely from marine aerosols.



in the significant negative correlation between Fe and TRANS (Fig. 2).

Biotic variables

Chlorophyll a

The concentration of CHLAU, which is widely used as a measure of phytoplankton biomass and trophic status (Vollenweider 1968), ranged from 0.1 to 20.4 $\mu g \cdot L^{-1}$ (Table 4). Thus, most of our study sites are oligotrophic according to Likens (1975) and Wetzel (1983). Above-average CHLAU levels were recorded in the forest–tundra (4.0 $\mu g \cdot L^{-1}$) and boreal forest lakes (2.5 $\mu g \cdot L^{-1}$) whereas average concentrations were considerably lower in arctic (1.4 $\mu g \cdot L^{-1}$) and alpine tundra (0.9 $\mu g \cdot L^{-1}$) lakes, reflecting both a latitudinal and altitudinal gradient in lake trophic status. This pattern is most likely associated with changing climatic and vegetational conditions and subsequent differences in nutrient input, length of growing season, and light regime (length of photoperiod), as well as other factors.

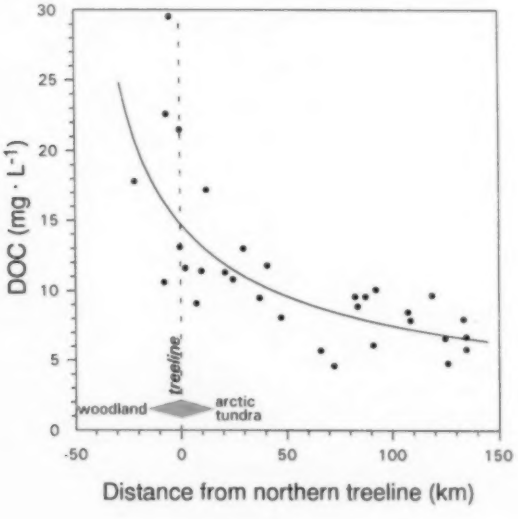
A significant CHLAU:TP relationship could be observed in our study lakes ($r = 0.62–0.64$, $P < 0.01$) (Table 3; Fig. 2), which corresponds to results of previous investigations of

© 1997 NRC Canada

Table 4 (*concluded*).

Na ($mg \cdot L^{-1}$)	K ($mg \cdot L^{-1}$)	Cl ($mg \cdot L^{-1}$)	CHLAU ($\mu g \cdot L^{-1}$)	CHLAC ($\mu g \cdot L^{-1}$)	POC ($\mu g \cdot L^{-1}$)	PN ($\mu g \cdot L^{-1}$)	Fe ($\mu g \cdot L^{-1}$)	Mn ($\mu g \cdot L^{-1}$)	TN:TP	POC:PN
4.01	1.01	1.9	2.0	ADL	360	52	265.0	14.0	31	7
2.07	0.98	2.5	1.5	ADL	480	72	160.0	20.0	35	7
3.71	0.39	0.3	0.8	ADL	308	52	56.9	35.0	26	6
3.40	0.58	0.4	1.1	0.8	396	56	35.2	11.0	42	7
0.34	0.10	0.4	0.9	0.6	664	96	664.0	22.0	26	7
0.34	0.39	0.6	1.1	ADL	608	84	127.0	17.0	54	7
0.70	0.45	0.3	0.8	0.6	616	88	196.0	160.0	19	7
0.24	0.14	0.2	0.9	0.7	252	40	37.4	11.0	31	6
0.24	0.10	0.2	0.1	0.1	158	26	5.9	2.0	13	5
187.00	29.90	75.2	20.4	18.1	3480	524	1660.0	160.0	103	9
4.01	1.15	2.0	1.4	0.6	492	72	80.6	14.0	40	7
8.82	1.90	7.3	2.1	1.2	714.1	102.9	210.7	23.5	45	7

Fig. 5. Plot of DOC concentrations versus distance from the northern treeline in a subset of 31 lakes from the Inuvik area and the Tuktoyaktuk Peninsula.



Yukon lakes (Shortreed and Stockner 1986). These results are not unexpected, given the large number of studies that document the strong correlation between chlorophyll *a* and TP in many North American lakes (e.g., Dillon and Rigler 1974; Kerekes 1975; Vollenweider 1976; Edmondson and Lehman 1981; Prepas and Trew 1983; Stockner and Shortreed 1985; Ostrofsky and Rigler 1987).

Principal components analysis

Because not all variables were measured in all study sites, a data set of 59 sample sites with 18 chemical (pH, COND, TPU, TPF, DOC, DIC, TKN, TN, SiO_2, SO_4, Ca, Na, K, Cl, POC, PN, Fe, Mn) and 4 physical variables (TEMP, TRANS, DEPTH, AREA) was used in the PCA (Fig. 2). Those variables with concentrations below or near the analytical limit of detection (i.e., NO_2, NO_3, NH_3, SRP) or variables that were not available for many of the lakes (i.e., DO) were excluded from the analysis. The biological variable CHLAU was given zero weight and included as a passive variable. The same was done with some of the geographic–environmental variables (i.e., LAT, ALT, FOREST, WOOD, TUNDRA, ALPINE), as the main purpose of this study was to illustrate and interpret the major patterns of variation within the physical and chemical characteristics of lakes. All passive variables were nevertheless included in the PCA ordination biplot (Fig. 2).

The total percentage of variation explained by the first two PCA axes was 56%, with eigenvalues of $\lambda_1 = 0.35$ and $\lambda_2 = 0.21$, respectively. Because the eigenvalues for the third and fourth PCA axes (0.11 and 0.08) were relatively low, they were not elaborated upon. The length of the solid arrows and their proximity to the axes show the relative weight of each variable in determining each axis (Ter Braak 1987). Thus, such variables as TRANS, DEPTH, AREA, Fe, Mn, TPF, and TPU are significantly correlated ($P < 0.05$) with axis 1 whereas the physical variables TEMP, LAT, and ALT are most strongly correlated with axis 2. The highly negative correlation between DEPTH and nutrient variables (including the trophic proxy CHLAU), as well as the strong positive correlation between DEPTH and TRANS, reflects the commonly observed inverse relationship between lake productivity and lake water depth – water transparency.

Two major groups of variables with high positive correlations (generally recognizable by the small angles between their biplot arrows) could be identified from the biplot, namely nutrient-related variables (i.e., TPU, POC, PN, TN, TKN, DOC) in the upper left quadrant of the ordination and those associated with ionic concentrations (i.e., COND, DIC, K, Na, Ca, SO_4), positioned in the upper right quadrant. Some of these variables were mutually redundant (e.g., TN and TKN) as demonstrated by their highly significant correlations (Table 3) and extremely small angles between their biplot arrows. Most variables were negatively correlated between these two major sets.

© 1997 NRC Canada

From the relative positioning of the sample scores, we infer that arctic and alpine tundra sites are physically and chemically very similar. Arctic and alpine tundra sites form a dense cloud of points in the central part of the lower two quadrants of the biplot, thereby indicating narrow ranges in physical and chemical conditions (Fig. 2). Boreal forest and forest–tundra sites, however, are not as distinct limnologically. As a result, their sample scores are more widely scattered in the upper two quadrants, reflecting conditions of comparatively wide variability (Fig. 2). The inclusion of a limited range of catchment characteristics (albeit run as passive variables) suggests that lakes in conifer-forested catchments generally tend to have higher concentrations of nutrients and major ions and that lakes with unforested catchments have considerably lower concentrations.

The first PCA axis effectively separated deep oligotrophic lakes with high water transparency, such as sites 7, 14, and 32, from shallow eutrophic sites with high [Fe], such as sites 56, 57, 49, and 24. The latter were generally strongly coloured with high nutrient concentrations, and mostly occurred in the forest–tundra near Inuvik.

From the eigenvalues and the placement of arrows related to nutrient and ionic variables, it was evident that both axes were relatively complex, thereby suggesting that there were several gradients of variation in the physical and chemical data. Both axes explained large proportions of the total variance, effectively separating "forested" sites characterized by either high concentrations in nutrients or major ions (or both), positioned in the upper two quadrants, from lakes located in areas of treeless vegetation (i.e., arctic and alpine tundra sites), positioned in the lower two quadrants.

Conclusions

The lakes distributed along the Yukon – Tuktoyaktuk Peninsula transect exhibit a wide range in water chemistry, thereby not only reflecting the strong climatic and vegetational gradients (both latitudinal and altitudinal) inherent in this data set, but also the complex nature of the geological and edaphic conditions of the study area. On the basis of their limnological characteristics, the 59 lakes we sampled can be separated into several distinct groups.

As expected, one of the most prominent spatial patterns arising from the data is the strong relationship between distance from the coast and [Cl] and [Na] in the lakes. Salts from atmospheric inputs (i.e., Cl^- and Na^+) are the dominant ions determining the chemistry of arctic coastal lakes on the Tuktoyaktuk Peninsula. Because permafrost acts as a seal and prevents subsurface drainage, these lakes are similar to other coastal lakes that are isolated from groundwater inputs. The chemistry of such coastal lakes is thus determined mainly by marine aerosols (e.g., Hammar 1989; Kling et al. 1992). In contrast with the input of sea salt to lakes near the coast, atmospheric inputs seem to have relatively little influence on the chemistry of inland lakes (Kling et al. 1992).

In the interior of the Yukon, lake waters are rich in HCO_3, Ca, and SO_4, and their proportions largely reflect the chemistry of the surrounding soils and bedrock, as well as catchment vegetation type. Measurements of pH, DIC, and Ca indicate that most surface waters were characterized by relatively high alkalinity and that acidic lakes were rare in the Yukon and northwestern Northwest Territories.

The strong latitudinal gradient exhibited in the concentrations of SiO_2 may be due to local bedrock geology, as well as differences in the external input of soil-derived elements such as Ca and SiO_2, which varies greatly between high- and low-latitude lakes. For example, the presence of permafrost distinguishes high-latitude lakes from lakes at lower latitudes. Permafrost has little direct effect on a lake but profoundly alters solute runoff from the lake's drainage, and may decrease the absolute amount of solute runoff as well by reducing the area of rock surface available for weathering (Welch and Legault 1986).

In general, the productivity is likely limited by the low levels of both inorganic nitrogen and inorganic phosphorus. However, all our study sites (including lakes on the Tuktoyaktuk Peninsula) showed markedly higher ionic compositions and nutrient levels than arctic lakes on the Canadian Shield (e.g., Pienitz et al. 1997). With respect to variables commonly used as indicators of trophic status (e.g., TP, TN, chlorophyll *a*), it appears that our study lakes from the southern and central part of the Yukon were slightly more productive than the 19 lakes studied by Shortreed and Stockner (1986), most of which were oligotrophic. This discrepancy may be due mainly to differences in lake area and depth, as our study sites were generally smaller and shallower. In the present study, lakes in boreal forest and forest–tundra regions are oligo- to meso-trophic whereas arctic and alpine tundra lakes are usually oligotrophic to ultraoligotrophic.

The organically bound nutrients in our freshwater lakes appeared to be closely associated with vegetational and climatic zones, with the most striking changes (i.e., sharp decreases in nutrients) usually occurring at the transition from forested areas to those located north of the treeline. Future limnological studies in these high-latitude regions should further elucidate some of these relationships.

Acknowledgements

This study was financed by the Natural Sciences and Engineering Research Council of Canada via a grant to J.P.S., a Canadian Government Scholarship to R.P., and Green Plan support on aquatic impacts of ultraviolet light to D.R.S.L. We thank Drs. R. Hall, A. Uutala, and I. Walker for help with the fieldwork. Fieldwork was supplemented by logistic support from the Polar Continental Shelf Project and the Northern Studies Training Program.

References

Allen, W.T.R. 1964. Break-up and freeze-up dates in Canada. Department of Transport, Meteorological Branch, Ottawa, Ont.

Anema, C., Hecky, R.E., Fee, E.J., Nernberg, D., and Guildford, S.J. 1990*a*. Water chemistry of some lakes and channels in the Mackenzie Delta and on the Tuktoyaktuk Peninsula, N.W.T., 1985. Can. Data Rep. Fish. Aquat. Sci. No. 726. (Available from Department of Fisheries and Oceans, Winnipeg, MB R3T 2N6, Canada.)

Anema, C., Hecky, R.E., Himmer, S., and Guildford, S.J. 1990*b*. Water chemistry of some lakes and channels in the Mackenzie Delta and on the Tuktoyaktuk Peninsula, N.W.T., 1986. Can. Data Rep. Fish. Aquat. Sci. No. 729. (Available from Department of Fisheries and Oceans, Winnipeg, MB R3T 2N6, Canada.)

© 1997 NRC Canada

Bello, R., and Smith, J.D. 1990. The effect of weather variability on the energy balance of a lake in the Hudson Bay Lowlands, Canada. Arct. Alp. Res. **22**: 98–107.

Bigras, S.C. 1990. Hydrological regime of lakes in the Mackenzie Delta, Northwest Territories, Canada. Arct. Alp. Res. **22**: 163–174.

Boer, G.J., McFarlane, N., Blanchet, J.-P., and Lazare, M. 1990. Greenhouse gas induced climatic change simulated with the CCC second generation GCM. *In* Application of the Canadian Climate Centre general circulation model output for regional climate impact studies: guidelines for users. Canadian Climate Centre, Atmospheric Environment Service, Downsview, Ont.

Cornwell, J.C. 1992. Cation export from Alaskan arctic watersheds. Hydrobiologia, **240**: 15–22.

Dillon, P.J., and Rigler, F.H. 1974. The phosphorus – chlorophyll relationship in lakes. Limnol. Oceanogr. **19**: 767–773.

Ecoregions Working Group. 1989. Ecoclimatic regions of Canada, first approximation. Ecological land classification series No. 23. Ecoregions Working Group, Environment Canada, Ottawa, Ont.

Edmondson, W.T., and Lehman, J.T. 1981. The effect of changes in the nutrient income on the condition of Lake Washington. Limnol. Oceanogr. **26**: 1–29.

Eilers, J.M., Landers, D.H., Newell, A.D., Mitch, M.E., Morrison, M., and Ford, J. 1993. Major ion chemistry of lakes on the Kenai Peninsula, Alaska. Can. J. Fish. Aquat. Sci. **50**: 816–826.

Ennis, G.L., Cinader, A., McIndoe, S., and Munon, T. 1982. An annotated bibliography and information summary on the fisheries resources of the Yukon River Basin in Canada. Can. Manuscr. Rep. Fish. Aquat. Sci. No. 1657.

Environment Canada. 1979. Analytical methods manual. Inland Waters Directorate, Water Quality Branch, Ottawa, Ont.

Environment Canada. 1985–1990. Yukon lake ice report data. Whitehorse Airport, Yukon Territory, Canada.

Fee, E.J., Hecky, R.E., Guildford, S.J., Anema, C., Mathew, D., and Hallard, K. 1988. Phytoplankton primary production and related limnological data for lakes and channels in the Mackenzie Delta and lakes on the Tuktoyaktuk Peninsula, N.W.T. Can. Tech. Rep. Fish. Aquat. Sci. No. 1614.

Hammar, J. 1989. Freshwater ecosystems of polar regions: vulnerable resources. Ambio, **18**: 6–22.

Hobbie, J.E. 1973. Arctic limnology: a review. *In* Alaskan arctic tundra. *Edited by* M.E. Britton. Technical paper No. 25. Arctic Institute of North America, Calgary, Alta. pp. 127–168.

Hobbie, J.E. (*Editor*). 1980. Limnology of tundra ponds. Dowden, Hutchinson & Ross, Inc., Stroudsburg, Pa.

Hobbie, J.E. 1984. Polar limnology. *In* Ecosystems of the World 23: lakes and reservoirs. *Edited by* F.B. Taub. Elsevier Science Publishers B.V., Amsterdam, The Netherlands. pp. 63–105.

Horler, A., Johnston, R.A.C., and Cronkite, G.M.W. 1983. An assessment of the fishery resource in 18 lakes within the Yukon Territory, Canada. Yukon River Basin Study Fisheries Work Group Project No. 3. Department of Fisheries and Oceans, Whitehorse, Y.T.

Howard, H.H., and Prescott, G.W. 1973. Seasonal variation of chemical parameters in Alaskan tundra lakes. Am. Midl. Nat. **90**: 154–164.

Kalff, J. 1968. Some physical and chemical characteristics of arctic fresh waters in Alaska and northwestern Canada. J. Fish. Res. Board Can. **25**: 2575–2587.

Kerekes, J.J. 1975. Limnological conditions in five small oligotrophic lakes in Terra Nova National Park, Newfoundland. J. Fish. Res. Board Can. **33**: 555–583.

Kling, G.W., Kipphut, G.W., and Miller, M.C. 1991. Arctic lakes and streams as gas conduits to the atmosphere: implications for tundra carbon budgets. Science (Washington, D.C.), **251**: 298–301.

Kling, G.W., O'Brien, W.J., Miller, M.C., and Hershey, A.E. 1992. The biogeochemistry and zoogeography of lakes and rivers in arctic Alaska. Hydrobiologia, **240**: 1–14.

Koivo, L.K., and Ritchie, J.C. 1978. Modern diatom assemblages from lake sediments in the boreal–arctic transition region near the Mackenzie Delta, N.W.T., Canada. Can. J. Bot. **56**: 1010–1020.

Lesack, L.F.W., Hecky, R.E., and Marsh, P. 1991. The influence of frequency and duration of flooding on the nutrient chemistry of Mackenzie Delta lakes. *In* Mackenzie Delta: Environmental Interactions and Implications of Development; Proceedings of the Workshop on the Mackenzie Delta, Saskatoon, Sask., October 17–18, 1989. *Edited by* P. Marsh and C.S.L. Ommanney. National Hydrology Research Institute, Environment Canada, Saskatoon, Sask. pp. 19–36.

Likens, G.E. 1975. Primary production of inland aquatic ecosystems. *In* The primary productivity of the biosphere. *Edited by* H. Lieth and R.W. Whittaker. Springer Verlag, New York. pp. 185–202.

Lindsey, C.C., Patalas, K., Bodaly, R.A., and Archibald, C.P. 1981. Glaciation and the physical, chemical, and biological limnology of Yukon lakes. Can. Tech. Rep. Fish. Aquat. Sci. No. 966.

Livingstone, D.A. 1966. Alaska, Yukon, Northwest Territories, and Greenland. *In* Limnology in North America. *Edited by* D.G. Frey. University of Wisconsin Press, Madison, Wis. pp. 559–574.

Luckman, B.H. 1989. Global change and the record of the past. GEOS, **18**: 1–8.

Manabe, S., and Wetherald, R.T. 1986. Reduction in summer soil wetness induced by an increase in atmospheric carbon dioxide. Science (Washington, D.C.), **232**: 626–628.

Marsh, P. 1989. Soil infiltration and snow-melt run-off in the Mackenzie Delta, N.W.T. *In* Proceedings of the 5th International Conference on Permafrost, Trondheim, Norway, August 2–5, 1988. Tapir Publishers, Trondheim, Norway. pp. 618–621.

Marsh, P., and Bigras, S.C. 1988. Evaporation from Mackenzie Delta lakes, N.W.T., Canada. Arct. Alp. Res. **20**: 220–229.

Maxwell, B. 1992. Arctic climate: potential for change under global warming. *In* Arctic ecosystems in a changing climate. *Edited by* F.S. Chapin, J.F. Reynolds, R.L. Jefferies, G.R. Shaver, J. Svoboda, and E.W. Chu. Academic Press, New York. pp. 11–34.

McNeely, R.N., Neimanis, V.P., and Dwyer, L. 1979. Water quality sourcebook: a guide to water quality parameters. Environment Canada, Inland Waters Directorate, Water Quality Branch, Ottawa, Ont.

Neilson, M.A., and Stevens, R.J.J. 1987. Spatial heterogeneity of nutrients and organic matter in Lake Ontario. Can. J. Fish. Aquat. Sci. **44**: 2192–2203.

Ostrofsky, M.L., and Rigler, F.H. 1987. Chlorophyll–phosphorus relationships for subarctic lakes in western Canada. Can. J. Fish. Aquat. Sci. **44**: 775–781.

Oswald, E.T., and Senyk, J.P. 1977. Ecoregions of Yukon Territory. Canadian Forestry Service, Environment Canada, Victoria, B.C.

Patalas, K. 1984. Mid-summer mixing depths of lakes of different latitudes. Verh. Int. Ver. Theor. Angew. Limnol. **22**: 97–102.

Pienitz, R. 1993. Paleoclimate proxy data inferred from freshwater diatoms from the Yukon and the Northwest Territories. Ph.D. thesis, Queen's University, Kingston, Ont.

Pienitz, R., Douglas, M.S.V., Smol, J.P., Huttunen, P., and Meriläinen, J. 1995*a*. Diatom, chrysophyte and protozoan distributions along a latitudinal transect in Fennoscandia. Ecography, **18**: 429–439.

Pienitz, R., Smol, J.P., and Birks, H.J.B. 1995*b*. Assessment of freshwater diatoms as quantitative indicators of past climatic change in the Yukon and Northwest Territories, Canada. J. Paleolimnol. **13**: 21–49.

Pienitz, R., Smol, J.P., and Lean, D.R.S. 1997. Physical and chemical limnology of 24 lakes located between Yellowknife and Cont-

© 1997 NRC Canada

woyto Lake, Northwest Territories (Canada). Can. J. Fish. Aquat. Sci. **54**: 347–358.

Prentki, R.T., Miller, M.C., Barsdate, R.J., Alexander, V., Kelley, J., and Coyne, P. 1980. Chemistry. *In* Limnology of tundra ponds. *Edited by* J.E. Hobbie. Dowden, Hutchinson & Ross, Inc., Stroudsburg, Pa. pp. 76–178.

Prepas, E.E., and Trew, D.O. 1983. Evaluation of the phosphorus-chlorophyll relationship for lakes off the Precambrian Shield in western Canada. Can. J. Fish. Aquat. Sci. **40**: 27–35.

Ritchie, J.C. 1977. The modern and late Quaternary vegetation of the Campbell–Dolomite Uplands near Inuvik, N.W.T., Canada. Ecol. Monogr. **47**: 401–423.

Ritchie, J.C. 1984. Past and present vegetation of the far northwest of Canada. University of Toronto Press, Downsview, Ont.

Roots, E.F. 1989. Climate change: high latitude regions. Clim. Change, **15**: 223–253.

Schindler, D.W., Welch, H.E., Kalff, J., Brunskill, G.J., and Kritsch, N. 1974. Physical and chemical limnology of Char Lake, Cornwallis Island (75°N lat.). J. Fish. Res. Board Can. **31**: 585–607.

Schindler, D.W., Beaty, K.G., Fee, E.J., Cruikshank, D.R., DeBruyn, E.R., Findlay, D.L., Linsey, G.A., Shearer, J.A., Stainton, M.P., and Turner, M.A. 1990. Effects of climatic warming on lakes of the central boreal forest. Science (Washington, D.C.), **250**: 967–970.

Schnitzer, M., and Desjardins, J.G. 1969. Chemical characteristics of a natural soil leachate from a humic podzol. Can. J. Soil Sci. **49**: 151–158.

Sheath, R.G. 1986. Seasonality of phytoplankton in northern tundra ponds. Hydrobiologia, **138**: 75–83.

Sheath, R.G., and Hellebust, J.A. 1978. Comparison of algae in the euplankton, tychoplankton, and periphyton of a tundra pond. Can. J. Bot. **56**: 1472–1483.

Shortreed, K.S., and Stockner, J.G. 1986. Trophic status of 19 subarctic lakes in the Yukon Territory. Can. J. Fish. Aquat. Sci. **43**: 797–805.

Stockner, J.G., and Shortreed, K.S. 1985. Whole-lake fertilization experiments in coastal British Columbia lakes: empirical relationships between nutrient inputs and phytoplankton biomass and production. Can. J. Fish. Aquat. Sci. **42**: 649–658.

Stumm, W., and Morgan, J.J. 1981. Aquatic chemistry. 2nd ed. John Wiley & Sons, New York.

Ter Braak, C.J.F. 1987. Unimodal models to relate species to environment. Agricultural Mathematics Group, Wageningen, The Netherlands.

Ter Braak, C.J.F. 1990*a*. CANOCO: a FORTRAN program for CANOnical Community Ordination. Microcomputer Power, Ithaca, N.Y.

Ter Braak, C.J.F. 1990*b*. Update notes: CANOCO version 3.10. Agricultural Mathematics Group, Wageningen, The Netherlands.

Van Tongeren, O.F.R., Van Liere, L., Gulati, R.D., Postema, G., and Boesewinkel-De Bruyn, P.J. 1992. Multivariate analysis of the plankton communities in the Loosdrecht lakes: relationship with the chemical and physical environment. Hydrobiologia, **233**: 105–117.

Vincent, W.F., and Ellis-Evans, J.C. (*Editors*). 1989. High latitude limnology. Hydrobiologia, **172**: 1–323.

Vollenweider, R.A. 1968. The scientific basis of lake and stream eutrophication, with particular reference to phosphorus and nitrogen as eutrophication factors. Techical report DAS/CSI/68. No. 27. Organisation for Economic Co-operation and Development, Paris. pp. 1–182.

Vollenweider, R.A. 1976. Advances in defining critical loading levels for phosphorus in lake eutrophication. Mem. Ist. Ital. Idrobiol. Dott Marco Marchi, **33**: 53–83.

Welch, H.E. 1991. Comparisons between lakes and seas during the arctic winter. Arct. Alp. Res. **23**: 11–23.

Welch, H.E., and Legault, J.A. 1986. Precipitation chemistry and chemical limnology of fertilized and natural lakes at Saqvaqjuac, N.W.T. Can. J. Fish. Aquat. Sci. **43**: 1104–1134.

Wetzel, R.G. 1983. Limnology. 2nd ed. Saunders College Publishing, Philadelphia, Pa.

Wilkinson, L. 1988. SYSTAT: the system for statistics. SYSTAT Inc., Evanston, Ill.

Zar, J.H. 1984. Biostatistical analysis. 2nd ed. Prentice-Hall Inc., Englewood Cliffs, N.J.

© 1997 NRC Canada

Physical and chemical limnology of 24 lakes located between Yellowknife and Contwoyto Lake, Northwest Territories (Canada)

Reinhard Pienitz, John P. Smol, and David R.S. Lean

Abstract: Data on the water chemistry and limnology of 24 lakes located between Yellowknife (62°27′N, 114°21′W) and Contwoyto Lake (65°30′N, 110°00′W) in the central Northwest Territories were examined using principal components analysis and other statistical techniques. The study sites were mostly shallow (Z_{max} = 2.5–25 m; mean = 8.2 m), nutrient-poor (3.4–12.7 μg total phosphorus/L; mean = 6.6 $\mu g \cdot L^{-1}$), low-alkalinity lakes. Water was typically low in solutes (specific conductance near 0–100 $\mu S \cdot cm^{-1}$), with slightly acidic to alkaline pH (6.2–8.9). Levels of all nutrients and major ions showed identical trends of decreasing concentrations with increasing latitude, with the highest concentrations generally in lakes with conifer-forested catchments in the south.

Résumé : Les données sur les caractéristiques chimiques et limnologiques de 24 lacs situés entre Yellowknife (62°27′N, 114°21′W) et le lac Contwoyto (65°30′N, 110°00′W) dans le centre des Territoires du Nord-Ouest (T.N.-O.) ont été examinées à l'aide de l'analyse des composantes principales. Les sites d'étude étaient pour la plupart des lacs peu profonds (Z_{max} = 2,5 à 25 m; moyenne = 8,2 m), pauvres en éléments nutritifs (3,4 à 12,7 $\mu g \cdot L^{-1}$ en phosphore totale, moyenne = 6,6 $\mu g \cdot L^{-1}$) et de faible alcalinité. De manière typique, l'eau était pauvre en solutés (conductivité électrique près de 0 à 100 $\mu S \cdot cm^{-1}$), et son pH variait de légèrement acide à alcalin (6,2 à 8,9). Les concentrations de tous les éléments nutritifs et des ions majeurs ont montré des tendances identiques, c.-à-d. qu'ils diminuaient avec l'augmentation de la latitude, les concentrations les plus élevées se trouvant généralement dans les lacs méridionaux avec forêts de conifères dans le bassin versant.
[Traduit par la Rédaction]

Introduction

This is the second of two papers (Pienitz et al. 1997) related to the chemical and limnological characteristics of lakes from the Canadian Arctic. Here we report on the results obtained during a survey of 24 lakes located between Yellowknife and Contwoyto Lake in the Northwest Territories. This survey was designed to examine the relationship between algal distributions and environmental variables (Pienitz and Smol 1993). The lakes are distributed across treeline along a latitudinal gradient (62°47′–65°27′N) that includes a strong vegetational gradient of boreal forests in the south to arctic tundra conditions in the north (Fig. 1). In addition to discussing the water chemistry and limnological characteristics of lakes within the study area, we also present a regional comparison of our results with data obtained from previous investigations in the area.

Received October 11, 1995. Accepted July 31, 1996.
J13106

R. Pienitz.[1] Centre d'études nordiques et département de géographie, Université Laval, Québec, QC G1K 7P4, Canada.
J.P. Smol. Paleoecological Environmental Assessment and Research Laboratory (PEARL), Department of Biology, Queen's University, Kingston, ON K7L 3N6, Canada.
D.R.S. Lean. Canada Centre for Inland Waters, National Water Research Institute, Box 5050, Burlington, ON L7R 4A6, Canada.

[1] Author to whom all correspondence should be addressed e-mail: reinhard.pienitz@cen.ulaval.ca

Although some water chemistry data have been reported for several lakes located in the vicinity of Yellowknife as part of assessments of their fishery potential (e.g., Healey and Woodall 1973*a*, 1973*b*; Wallace and Hardin 1975; Falk 1979*a*, 1979*b*; Moore 1979, 1981; Moore et al. 1979; Patalas 1984), most of the data are restricted to single observations of limnological variables and very few complete ion analyses. Despite the relatively large number of these earlier studies, information on the limnological characteristics of lakes in more remote areas of the Northwest Territories remains sparse.

The objectives of the present paper are the same as those outlined in Pienitz et al. (1997), namely to describe the physical and chemical characteristics of our study sites. We also attempted to identify patterns in limnological conditions and to relate these patterns to watershed characteristics. For this purpose, we applied principal components analysis (PCA), an ordination technique that is useful for detecting the internal structure in data sets including a large number of interrelated variables (Van Tongeren et al. 1992). This study should provide reference data for evaluating future changes in the limnological conditions of northern lakes.

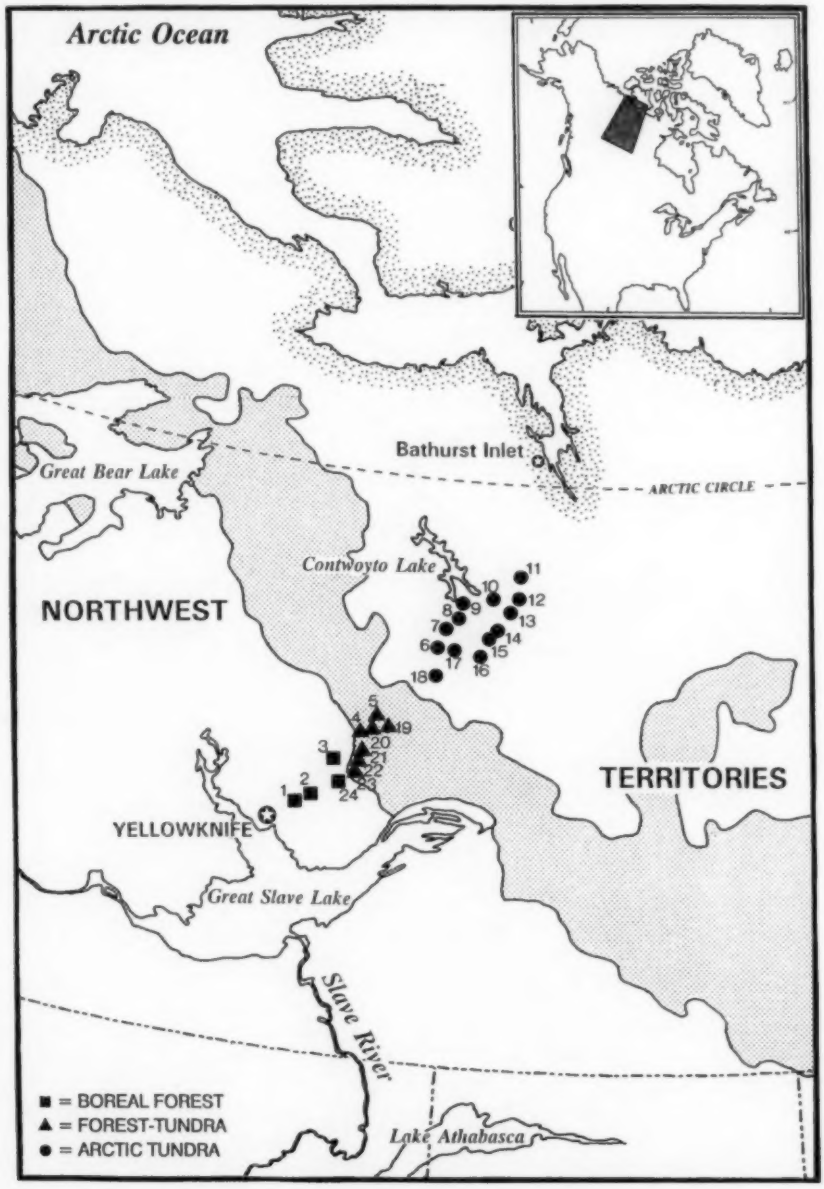
Description of study area

Regional climate and hydrology

The more than 500 km long south–north transect that makes up the study area spans three major ecoclimatic provinces (Boreal, Subarctic, Arctic), including the boundaries between high boreal, low subarctic, high subarctic, and low arctic ecoclimatic regions (Ecoregions Working Group

© 1997 NRC Canada

Fig. 1. Location of the 24 study sites and major towns in the central Northwest Territories.



1989). It crosses the steep climatic gradients that characterize the treeline region (summarized in Table 1). Sharp declines in mean annual precipitation and temperature are due to large differences in climate that prevail south and north of the mean summer position of the Arctic Front (Bryson 1966) as well as changes in albedo from forested to treeless areas (Hare and Ritchie 1972).

Lakes within the boreal forest near Yellowknife are usually ice covered by late October and break up in late May to early June (Environment Canada 1985–1991). In small- to medium-sized tundra lakes (50–250 ha), such as sites 6–18, the ice cover forms about 2 weeks earlier and usually remains until the end of June (ice reports, Yellowknife weather station). Thus, the southern lakes have nearly 5 months of open water and a growing season at least 4–5 weeks longer than those in the north. On small lakes, about 1–1.5 m of ice forms (Healey and Woodall 1973*a*; G.M. MacDonald, UCLA, personal communication). In 1991, some of the larger lakes in the northern part of the study area near Contwoyto Lake were still partly ice covered on July 12.

© 1997 NRC Canada

Table 1. Climate data from the study area.

	Yellowknife (62°27′N, 114°21′W)	Contwoyto Lake (65°30′N, 110°00′W)
January mean daily air temperature (°C)	−28.8	−32.1
July mean daily air temperature (°C)	16.3	9.7
Mean annual daily temperature (°C)	−5.4	−12.0
Mean annual precipitation (mm)	266.7	251.3
Growing degree-days above 5°C	1027.1	339.8

The study area belongs to the Precambrian Shield (Canadian Shield) geological region, which is characterized by rolling terrain with gentle relief. The Yellowknife area lies within the Slave Structural Province, which is an Archean craton comprising numerous supracrustal belts and extensive gneissic–granitoid plutons (Padgham and Fyson 1992). Supracrustal rocks are characterized by a predominance of sedimentary over volcanic rocks and are collectively termed the Yellowknife Supergroup (Henderson 1970, 1981). According to Padgham and Fyson (1992), the Yellowknife Supergroup consists of three distinct assemblages of which the most widespread and abundant consists of metasedimentary turbidites. The scouring action of inland glaciers left ridge tops and steeper slopes nearly till free and lower slopes and valleys with a shallow overburden of coarse glacial till. Most of the numerous lakes in the area occupy ice-scour depressions in bedrock (Livingstone 1966) whereas extensive muskegs and bogs result from poor drainage in these regions of low relief. Soil conditions are influenced by permafrost, which is continuous north of the treeline and discontinuous throughout the rest of the region. For more detailed information on the geology and glacial history of the study area, refer to Pienitz (1993).

Vegetation

Boreal forest, forest–tundra, and tundra comprise the vegetation of the study area. The forest is characterized by a closed or continuous canopy, with *Picea mariana*, *Picea glauca*, *Larix laricina*, *Pinus banksiana*, *Betula papyrifera*, *Populus tremuloides*, and *Populus balsamifera*. Poorly drained soils are extensive and are usually dominated by black spruce (Rowe 1972).

Moving northward to the forest–tundra, trees are restricted to areas with favourable local conditions of slope, aspect, and edaphic conditions. Forest patches are dominated by *Picea mariana*, but may include *Picea glauca* and *L. laricina*. Open areas are occupied by *Alnus crispa*, *Betula glandulosa*, *Salix* spp., and diverse sedges, grasses, herbs, mosses, and lichens. Peatlands often include *Picea mariana* with an understory of herbs and shrubs, including *A. crispa*, *B. glandulosa*, *Myrica gale*, sedges, heaths, and *Sphagnum* spp.

Further north the forest disappears, although lone individuals or small stands of black spruce krummholz can be found well beyond the mapped limits of the forest–tundra. The tundra vegetation includes *A. crispa*, *B. glandulosa*, *Salix* spp., grasses, sedges, herbs, as well as mosses and lichens. Vegetation in low-lying areas is dominated by members of the Cyperaceae, especially *Carex* and *Eriophorum*. On upland sites, characterized by exposed bedrock, tundra herbs and lichens are common.

Materials and methods

The study sites were sampled on July 12, 1991, using a helicopter equipped with pontoons. All 24 lakes are unnamed, and therefore are referred to as sites 1–24. Full details of field sampling methods and analytical protocols are provided in Pienitz et al. (1997) and the *Analytical Methods Manual* (Environment Canada 1979). Water chemistry analyses were performed by the National Water Research Institute (Burlington, Ont.). For logistical reasons, no measurements of O_2 were carried out.

Data analysis

The physical and chemical characteristics of the 24 sampling sites are shown in Tables 2 and 3, along with their respective minimum, maximum, median, and mean values. These data included water chemistry (pH, specific conductance (COND), total unfiltered phosphorus (TPU), total filtered phosphorus (TPF), soluble reactive phosphorus (SRP), nitrite (NO_2), nitrate (NO_3), ammonia (NH_3), total Kjeldahl nitrogen (TKN), total nitrogen (TN), particulate nitrogen (PN), particulate organic carbon (POC), dissolved organic carbon (DOC), dissolved inorganic carbon (DIC), silica (SiO_2), chloride (Cl), sulphate (SO_4), sodium (Na), potassium (K), calcium (Ca), iron (Fe), and manganese (Mn)), Secchi depth (TRANS), surface water temperature (TEMP), maximum depth (DEPTH), lake area (AREA), altitude (ALT), latitude (LAT), and longitude (LONG), shortest distance from northern treeline (DIST), as well as concentrations of uncorrected chlorophyll *a* (CHLAU) and corrected chlorophyll *a* (CHLAC). The above abbreviations in parentheses correspond to the labels used for all 36 variables in the figures and tables.

In general, criteria for initial data screening and subsequent data transformation followed those outlined in Pienitz et al. (1997). The variables DIC, Ca, Na, K, Cl, Fe, and COND were positively skewed, and therefore were $\ln(x + 1)$ transformed to normalize distributions (Zar 1984), where x is the environmental variable under consideration.

Patterns of correlation (discussed below) between the variables were identified from a Pearson correlation matrix (Table 4) with Bonferroni-adjusted significance values (Wilkinson 1988). We used PCA, available on the computer program CANOCO 3.1 (Ter Braak 1990*a*, 1990*b*), to summarize the major patterns of variation within the data and to ordinate the lakes with respect to their chemical and limnological characteristics. Catchment vegetation (FOREST, closed-canopy boreal forest; WOOD, forest–tundra and lichen–woodland; TUNDRA, arctic tundra) was included as binary codes (value 0 or 1) in the PCA.

A PCA correlation biplot was derived from a PCA of the physical, chemical, and environmental data (Fig. 2). The five environmental variables LAT, ALT, FOREST, WOOD, and TUNDRA, as well as the biological variable CHLAU, were included only as passive

© 1997 NRC Canada

Table 2. Field data collected in 1991 for the 24 lakes.

Lake No.	LAT (°N)	LONG (°W)	ALT (m)	AREA (ha)	Z_{max} (m)	pH	TEMP (°C)	COND ($\mu S \cdot cm^{-1}$)	TRANS (m)	DIST (km)
Boreal forest										
1	62.47	113.40	274	180.3	13.0	8.5	14.5	100	4.8	–120.0
2	62.50	113.28	305	83.4	25.0	8.0	14.5	48	3.3	–112.5
3	63.18	112.55	396	223.7	10.0	8.7	13.0	30	7.5	–55.0
24	63.06	112.42	380	301.1	9.0	8.9	14.0	32	3.0	–68.0
Forest–tundra										
4	63.33	112.22	427	113.8	2.5	8.2	12.0	12	2.0	–15.0
5	63.43	112.05	427	79.2	3.0	7.7	12.0	11	**3.0**	12.5
19	63.35	111.54	450	130.5	7.0	8.2	13.0	0	2.5	12.5
20	63.39	112.08	427	288.4	6.0	8.2	11.5	0	2.5	5.0
21	63.22	112.23	427	97.5	2.5	7.5	14.0	20	2.0	–30.0
22	63.16	112.28	389	283.8	10.0	7.8	13.0	20	2.8	–47.5
23	63.13	112.29	396	98.2	5.5	8.8	14.0	22	2.5	–52.5
Arctic tundra										
6	64.37	110.43	457	254.4	8.0	8.0	10.0	10	4.0	130.0
7	64.51	110.25	471	488.5	5.0	8.1	8.0	10	**5.0**	157.5
8	64.55	110.12	457	111.6	6.0	7.9	9.0	10	5.5	170.0
9	65.06	110.13	488	262.5	8.0	8.2	8.0	8	5.0	185.0
10	65.13	109.11	457	175.6	20.0	8.6	7.5	9	6.5	220.0
11	65.27	108.33	427	263.1	5.0	8.4	9.0	8	4.5	262.5
12	65.16	108.29	396	421.1	15.0	8.0	8.0	10	5.0	242.5
13	65.05	108.30	442	244.5	8.5	6.3	8.5	10	6.5	227.5
14	64.50	109.11	440	68.3	3.5	7.4	8.0	8	2.3	195.0
15	64.47	109.13	440	52.7	3.5	7.5	9.5	8	**3.5**	190.0
16	64.31	109.38	430	501.7	7.0	6.2	9.5	8	4.3	155.0
17	64.35	110.17	457	119.8	6.0	6.2	10.5	8	3.5	142.5
18	64.18	110.38	459	317.1	8.0	8.0	10.5	8	3.5	107.5
Minimum	62.47	108.29	274	52.7	2.5	6.2	7.5	0	2.0	–120.0
Maximum	65.27	113.40	488	501.7	25.0	8.9	14.5	100	7.5	262.5
Median			430	223.7	7.0	8.0	10.5	10	3.5	130.0
Mean			421.6	215.0	8.2	7.9	10.9	17.1	3.9	79.8

Note: TRANS, Secchi disk transparency (bold numbers are readings where TRANS = Z_{max}); DIST, shortest distance from northern treeline (defined as 80% tree cover).

variables (represented by arrows with broken lines in Fig. 2). In addition, the five variables NO_2, NO_3, NH_3, SRP, and Mn were excluded from the analysis because their concentrations were frequently below or near the analytical detection limit. In all cases, the criterion level for significance was $P \leq 0.05$.

Results and discussion

The correlation matrix of environmental variables shows that many of the variables were highly correlated (Table 4). As expected, altitude and latitude were both negatively correlated with temperature, conductivity, major ions, and macronutrients (TPU, TN). Lake depth had little correlation with lake water chemistry. DOC was highly correlated with LAT, TEMP, and nutrients whereas DIC was highly correlated with Ca ($r = 0.99$, $P < 0.01$), as well as Cl, Na, and K. Both DOC and DIC were highly correlated with catchment vegetation. The inclusion of a limited range of catchment characteristics (albeit run as passive variables in the PCA) suggested that levels of all organic forms of nutrients and major ions were generally highest in lakes with conifer-forested catchments, with concentrations decreasing as distance from the northern treeline increased.

Physical and geographic–environmental variables

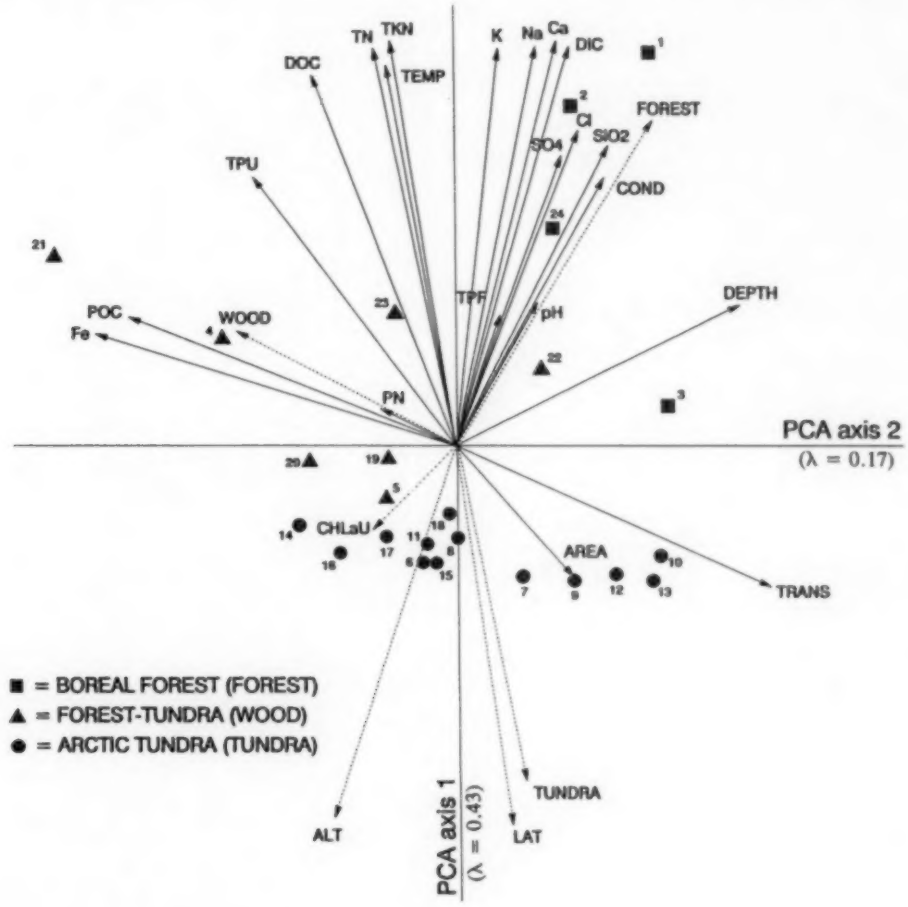
A regular shoreline, a circular outline, and the absence of major inflows were the main criteria we used to select lakes. The altitudinal gradient was comparatively small in this calibration set of lakes, since lake elevation was quite uniform, ranging only from 274 to 488 m above sea level (Table 2).

Lake surface area varied between 53 (15) and 502 ha (16), the average area being 215 ha (Table 2). Thus, we were dealing mainly with small shallow lakes with maximum depths ranging from 2.5 (4, 21) to 25 m (2).

Summer surface water temperatures were low and showed a strong latitudinal gradient, with a particularly sharp drop in temperature in lakes located north of the treeline. Temperatures ranged from 14.5°C (1, 2) in the boreal forest to 7.5°C (10) in the tundra and averaged 10.9°C for all lakes. The aver-

© 1997 NRC Canada

Fig. 2. PCA of the physical, chemical, and environmental variables in the 24-lake data set. Passive variables are represented by arrows with broken lines.



age water temperature was 14.0°C in boreal forest and 12.8°C in forest–tundra lakes whereas it was considerably lower in arctic tundra lakes (8.9°C). A strong relationship exists between thermal features (i.e., lake surface water temperature) of lakes and climatic gradients (represented by latitude in Fig. 3). The temperature pattern observed in our lake transect was consistent with results reported from studies of thermal regimes in North American lakes (e.g., Shuter et al. 1983; Robertson 1989; Hobbie et al. 1995).

Owing to logistic constraints of sampling from a helicopter, we could not obtain temperature profiles along this transect. Information available from previous investigations (Healey and Woodall 1973*a*, 1973*b*; Falk 1979*a*, 1979*b*) indicates that most lakes in the Yellowknife area stratify during the summer months. The unusually low water temperatures show that heat income during the open-water season of 1991 was minimal owing to a very cold and rainy summer.

The lowest water transparencies were observed in lakes surrounded by peatlands (most of which were found in the forest–tundra), where Secchi depths were commonly between 2.0 and 3.0 m (e.g., sites 4, 5, 21). The latter may be due to high loadings of humic compounds from terrestrial vegetation and soils in the drainage basin, as demonstrated by the relatively high concentrations of DOC, POC, and Fe (Table 3) in these coloured lakes.

Chemical variables

pH

Measurements of pH exhibited a high variability for lakes located in an area of relatively homogeneous geological and edaphic conditions, ranging from 6.2 to 8.9 (Table 2). All 24 lakes lie on granitoid basement rock and receive most of their drainage from similar rocks, although there are extensive outcrops of metasedimentary rocks (i.e., mainly carbonate-containing greywacke and mudstone turbidites belonging to the Burwash Formation of the Yellowknife Supergroup) in the Yellowknife area (Henderson 1981; Padgham and Fyson 1992), where pH values tended to be higher. Likewise, locally elevated pH values may also be related to isolated, narrow

© 1997 NRC Canada

Table 3. Water chemistry data (1991) for the 24 lakes.

Lake No.	TPU ($\mu g \cdot L^{-1}$)	TPF ($\mu g \cdot L^{-1}$)	SRP ($\mu g \cdot L^{-1}$)	NO_2 ($\mu g \cdot L^{-1}$)	NO_3 ($\mu g \cdot L^{-1}$)	NH_3 ($\mu g \cdot L^{-1}$)	DOC ($mg \cdot L^{-1}$)	DIC ($mg \cdot L^{-1}$)	TKN ($\mu g \cdot L^{-1}$)	TN ($\mu g \cdot L^{-1}$)	SiO_2 ($mg \cdot L^{-1}$)	SO_4 ($mg \cdot L^{-1}$)
Boreal forest												
1	9.5	5.2	0.6	2.0	ADL	ADL	5.6	12.1	352	425	0.41	2.1
2	9.6	7.6	0.8	2.0	ADL	13.0	8.5	5.6	371	478	0.92	2.3
3	3.9	2.3	0.6	2.0	ADL	6.0	4.7	3.5	176	247	0.61	0.6
24	9.2	5.8	0.8	2.0	14.0	13.0	8.1	3.1	282	344	0.77	1.4
Forest–tundra												
4	9.6	4.7	1.0	3.0	ADL	12.0	8.7	0.4	325	389	0.32	1.4
5	3.6	2.7	0.5	ADL	ADL	9.0	4.0	0.6	178	242	0.24	0.3
19	6.1	3.2	1.3	3.0	ADL	6.0	4.9	0.6	173	260	0.34	1.2
20	5.2	4.0	0.6	1.0	ADL	12.0	4.5	1.1	183	264	0.13	0.9
21	12.7	7.8	1.1	3.0	ADL	6.0	9.1	1.7	321	429	0.16	0.8
22	6.5	5.3	0.7	3.0	ADL	8.0	5.7	2.1	203	267	0.64	1.4
23	5.9	5.7	0.8	2.0	ADL	9.0	6.4	2.4	270	375	0.38	1.0
Arctic tundra												
6	7.7	2.8	0.4	1.0	ADL	11.0	1.7	0.5	63	135	0.08	0.5
7	5.5	9.0	0.4	1.0	ADL	6.0	2.4	0.3	102	212	0.10	0.4
8	6.3	3.3	0.5	ADL	ADL	11.0	2.0	0.2	150	266	0.26	1.0
9	4.5	8.7	0.5	3.0	ADL	5.0	2.2	0.2	141	211	0.19	0.4
10	3.4	2.2	0.5	1.0	ADL	ADL	2.7	0.2	126	240	0.18	0.9
11	9.7	3.0	0.5	1.0	ADL	12.0	2.6	0.3	147	211	0.27	0.3
12	3.6	3.1	0.4	1.0	ADL	8.0	2.0	0.1	105	161	0.21	1.5
13	4.6	9.2	0.4	2.0	ADL	5.0	1.6	0.5	88	147	0.26	1.4
14	7.3	2.7	0.6	1.0	ADL	8.0	3.2	0.2	160	233	0.19	1.0
15	5.5	1.8	0.5	1.0	ADL	9.0	2.5	0.3	123	164	0.18	0.7
16	7.7	2.9	0.6	3.0	ADL	ADL	3.2	0.2	32	146	0.19	1.0
17	4.0	2.9	0.5	2.0	ADL	ADL	2.4	0.3	115	222	0.33	0.9
18	6.7	2.3	0.6	3.0	ADL	6.0	3.8	0.1	157	213	0.29	1.2
Minimum	3.4	1.8	0.4	1.0	ADL	5.0	1.6	0.1	32	135	0.08	0.3
Maximum	12.7	9.2	1.3	3.0	14.0	13.0	9.1	12.1	371	478	0.92	2.3
Median	6.3	3.3	0.6	2.0		8.0	3.8	0.5	160	242	0.26	1.0
Mean	6.6	4.5	0.6	1.9		8.1	4.3	1.5	181.0	261.7	0.32	1.0

Note: ADL, at detection limit; TPU, total phosphorus (unfiltered); TPF, total phosphorus (filtered); SRP, soluble reactive phosphorus; CHLAU, chlorophyll *a* (uncorrected); CHLAC, chlorophyll *a* (corrected).

belts of volcanic rocks of the Supergroup, which include both mafic–felsic series and calc–alkaline intermediate series (e.g., Frith 1987; Cunningham and Lambert 1989).

In general, fresh waters within Shield areas in the Northwest Territories are known to be highly sensitive to acidification because of their low buffering capacity (Tibbatts et al. 1987). Previous surveys of lakes in the area between Yellowknife and Contwoyto Lake yielded consistently low and uniform pH values between 6.3 and 7.3 (e.g., Moore 1979; Tibbatts et al. 1987). However, Healey and Woodall (1973*b*) and Falk (1979*a*) reported summer pH values ranging from 7.4 to 9.3 and from 6.6 to 7.9, respectively, for several lakes in the Yellowknife area.

Conductivity and major ions

In general, the surface waters of our study sites were very dilute. Conductivity was consistently low and ranged from 0 to 100 $\mu S \cdot cm^{-1}$ (mean = 17.1 $\mu S \cdot cm^{-1}$) (Table 2), which was similar but slightly lower than previously reported values (10–350 $\mu S \cdot cm^{-1}$) for arctic lakes on Precambrian rock (Rawson 1960; Healey and Woodall 1973*a*; Hobbie 1984; Welch 1985; Welch and Legault 1986). Most variability seemed to be related to local vegetation; values in boreal forest lakes greatly exceeded those recorded in forest–tundra and arctic tundra lakes.

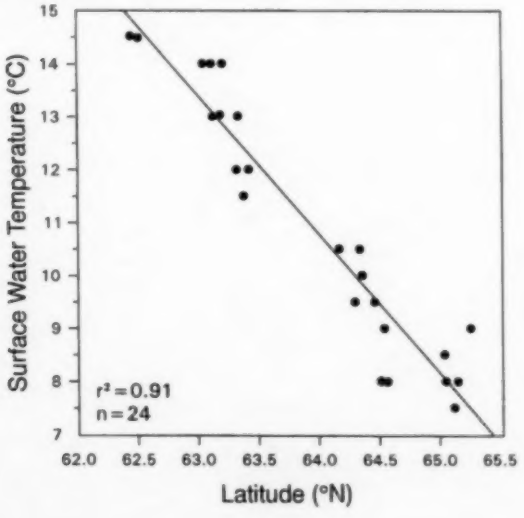
The concentrations of major ions were also consistently low. The data obtained for SO_4, Ca, Na, K, and Cl yielded identical trends of decreasing concentrations from boreal forest to tundra sites. The relative concentration of cations in all lakes was Ca > Na > K (Table 3). All but one variable (SO_4) tended to be highly positively correlated with each other ($P < 0.05$), as well as with DIC (Table 4). In lakes along the Yellowknife – Contwoyto Lake transect, where granitic–gneissic rocks predominate, Ca levels were very low, with an average summer concentration of 2.2 $mg \cdot L^{-1}$ for all lakes (Table 3). This value is much lower than that reported from Yukon lakes (mean = 19.7 $mg \cdot L^{-1}$; Pienitz et al. 1997), where waters from regions with deposits of limestone, dolo-

© 1997 NRC Canada

Table 3 (*concluded*).

Ca ($mg \cdot L^{-1}$)	Na ($mg \cdot L^{-1}$)	K ($mg \cdot L^{-1}$)	Cl ($mg \cdot L^{-1}$)	CHLAU ($\mu g \cdot L^{-1}$)	CHLAC ($\mu g \cdot L^{-1}$)	POC ($\mu g \cdot L^{-1}$)	PN ($\mu g \cdot L^{-1}$)	Fe ($\mu g \cdot L^{-1}$)	Mn ($\mu g \cdot L^{-1}$)	TN:TP	POC:PN
13.5	4.66	1.95	4.8	0.8	0.5	488	60	36.6	4.0	45	8
6.3	1.82	1.16	1.0	0.8	0.6	641	94	31.2	2.0	50	7
5.1	0.81	0.58	0.7	0.4	0.2	390	58	17.5	3.0	63	7
4.9	1.55	0.67	2.1	0.6	0.4	525	46	23.9	ADL	37	11
1.2	0.79	0.51	0.7	0.8	0.6	532	50	178.0	8.0	41	11
1.1	0.67	0.40	0.6	0.8	0.7	483	52	43.6	4.0	67	9
1.3	0.67	0.42	0.8	0.7	0.3	580	73	30.1	ADL	43	8
1.6	0.77	0.60	0.7	0.7	0.4	899	69	44.3	3.0	51	13
2.6	0.93	0.70	0.8	1.4	0.9	1080	94	210.0	12.0	34	11
2.9	0.79	0.68	0.6	0.6	0.2	465	50	18.7	ADL	41	9
3.4	0.83	0.83	0.6	0.8	0.5	592	92	17.0	10.0	64	6
0.7	0.44	0.46	0.6	0.5	0.1	595	60	49.3	ADL	18	10
0.6	0.46	0.37	0.7	1.2	0.9	484	98	16.1	ADL	39	5
0.5	0.42	0.34	0.5	0.8	0.2	550	104	27.2	ADL	42	5
0.6	0.38	0.33	0.6	1.3	1.1	375	56	10.2	ADL	47	7
0.7	0.42	0.34	0.8	0.8	0.1	387	102	10.3	ADL	71	4
0.7	0.46	0.30	0.6	1.0	0.7	521	52	34.3	ADL	22	10
0.7	0.44	0.35	0.6	1.1	0.4	484	44	17.1	ADL	45	11
1.2	0.36	0.27	0.5	0.9	0.8	354	46	7.5	ADL	32	8
0.7	0.38	0.40	0.5	1.0	0.8	578	61	134.0	ADL	32	9
0.7	0.42	0.38	0.6	1.8	1.4	528	29	21.7	ADL	30	18
0.7	0.46	0.47	0.7	0.7	0.6	598	100	53.1	26.0	19	6
0.6	0.46	0.44	0.5	0.6	0.2	549	94	51.3	4.0	56	6
0.8	0.40	0.34	0.8	0.7	0.3	577	42	33.5	2.0	32	14
0.5	0.36	0.27	0.5	0.4	0.1	354	29	7.5	2.0	18	4
13.5	4.66	1.95	4.8	1.8	1.4	1080	104	210.0	26.0	71	18
1.1	0.46	0.44	0.7	0.8	0.5	532	60	31.2	4.0	42	9
2.2	0.82	0.55	0.9	0.9	0.5	552.3	67.8	46.5	4.3	43	9

Fig. 3. Plot of surface water temperature versus latitude in the 24-lake data set.



mite, and gypsum are more prevalent. Waters with [Ca] ≤ 10 $mg \cdot L^{-1}$ are usually oligotrophic. The higher levels of Ca in lakes near Yellowknife (sites 1–3, 21–24) may be related to the more widespread sedimentary rocks of the Burwash Formation (see above).

Nutrients and carbon

Patterns in nutrient concentrations paralleled our observations of ionic concentrations, with values decreasing from forested to barren landscapes. Concentrations of dissolved nutrients were generally low; many variables (SRP, NO_3, NO_2, NH_3, and SiO_2) were near or below analytical detection limit, particularly in the arctic tundra lakes. NO_3 levels were consistently below the analytical detection limit (11.0 $\mu g \cdot L^{-1}$) in all but one lake (24). The concentrations of SiO_2 were very low, ranging from 0.08 to 0.92 $mg \cdot L^{-1}$ (Table 3).

The concentrations of both DIC and DOC displayed strong latitudinal gradients, with concentrations decreasing as latitude increased. This strong relationship was also demonstrated in the highly significant negative correlation between DIC–DOC and LAT (Table 4; Fig. 2). The concentrations of DIC ranged from 0.1 to 12.1 $mg \cdot L^{-1}$ and were generally

© 1997 NRC Canada

Table 4. Pearson correlation matrix for environmental variables (following transformations) measured in the 24 lakes included in the PCA.

	LAT	LONG	ALT	DEPTH	AREA	TEMP	TRANS	COND	pH	ZOOPL	CHLAU	CHLAC	TPU	TPF	SRP	NO_2	NO_3
LONG	−0.96**																
ALT	0.69	−0.59															
DEPTH	−0.11	0.11	−0.52														
AREA	0.31	−0.29	0.13	0.09													
TEMP	−0.95**	0.91**	−0.68	0.09	−0.34												
TRANS	0.46	−0.38	0.08	0.38	0.32	−0.45											
COND	−0.36	0.36	−0.65	0.35	−0.10	0.38	0.20										
pH	−0.32	0.41	−0.20	0.22	−0.08	0.34	−0.06	−0.02									
ZOOPL	0.01	−0.05	−0.05	0.03	0.30	0.02	0.22	0.03	0.05								
CHLAU	0.35	−0.37	0.21	−0.24	−0.13	−0.34	−0.09	−0.04	−0.18	−0.41							
CHLAC	0.17	−0.21	0.11	−0.35	−0.11	−0.20	−0.15	0.03	−0.28	−0.40	0.86**						
TPU	−0.42	0.36	−0.40	−0.09	−0.13	0.47	−0.47	0.35	0.02	−0.17	0.09	0.15					
TPF	−0.14	0.21	−0.13	0.09	0.14	0.14	0.01	0.27	−0.05	−0.24	0.26	0.40	0.25				
SRP	−0.62	0.56	−0.20	−0.10	−0.36	0.66	−0.60	−0.10	0.31	−0.21	−0.08	−0.05	0.52	0.12			
NO_2	−0.37	0.36	−0.07	−0.05	0.06	0.43	−0.27	0.09	−0.12	0.04	−0.15	−0.01	0.33	0.30	0.62		
NO_3	−0.22	0.21	−0.18	0.03	0.14	0.27	−0.13	0.22	0.38	−0.03	−0.18	−0.09	0.23	0.12	0.15	0.03	
NH_3	−0.25	0.24	−0.23	0.00	−0.20	0.25	−0.39	0.01	0.34	−0.24	−0.12	−0.06	0.36	−0.09	0.11	−0.33	0.36
TKN	−0.79**	0.79**	−0.70	0.19	−0.47	0.80**	−0.43	0.50	0.40	−0.17	−0.04	0.03	0.61	0.32	0.61	0.31	0.23
TN	−0.78**	0.79**	−0.66	0.23	−0.46	0.77**	−0.40	0.49	0.38	−0.15	−0.07	−0.03	0.60	0.35	0.63	0.30	0.18
PN	−0.06	0.10	0.07	0.18	−0.02	0.02	0.06	−0.01	−0.02	0.03	−0.14	−0.24	0.07	0.16	0.14	−0.01	−0.20
POC	−0.36	0.31	−0.05	−0.25	−0.16	0.37	−0.61	−0.21	0.01	0.08	0.15	0.07	0.56	0.06	0.47	0.12	−0.04
DOC	−0.83**	0.80**	−0.56	0.09	−0.31	0.84**	−0.55	0.38	0.35	−0.08	−0.13	−0.02	0.65	0.29	0.77**	0.49	0.34
DIC	−0.82**	0.79**	−0.89**	0.39	−0.20	0.82**	−0.05	0.65	0.53	0.08	−0.27	−0.14	0.39	0.28	0.30	0.18	0.25
SiO_2	−0.63	0.59	−0.68	0.51	−0.16	0.66	−0.06	0.53	0.25	−0.11	−0.41	−0.26	0.23	0.17	0.33	0.30	0.45
Cl	−0.53	0.50	−0.72*	0.30	0.02	0.50	0.01	0.52	0.33	0.04	−0.14	−0.08	0.40	0.14	0.16	0.12	0.40
SO_4	−0.49	0.36	−0.74*	0.59	−0.09	0.45	−0.14	0.39	0.04	0.04	0.22	−0.22	0.31	0.20	0.31	0.29	0.15
Na	−0.76**	0.73*	−0.89**	0.36	−0.17	0.74*	−0.13	0.60	0.31	−0.03	−0.19	−0.09	0.48	0.23	0.31	0.17	0.27
K	−0.80**	0.75*	−0.89**	0.36	−0.20	0.76*	−0.20	0.61	0.24	0.07	−0.19	−0.10	0.47	0.23	0.31	0.20	0.10
Ca	−0.82**	0.78**	−0.89**	0.39	−0.18	0.83**	−0.06	0.65	0.36	0.13	−0.28	−0.14	0.39	0.27	0.35	0.25	0.29
Fe	−0.32	0.25	−0.04	−0.43	−0.31	0.27	−0.66	−0.03	−0.22	−0.08	0.00	0.06	0.63	−0.20	0.45	0.17	−0.07
Mn	−0.28	0.22	−0.09	−0.30	0.05	0.30	−0.29	0.13	−0.23	0.34	−0.03	0.10	0.34	0.03	0.34	0.43	−0.12
DIST	0.99**	−0.98**	0.67	−0.14	0.27	−0.96**	0.40	−0.40	−0.39	−0.02	0.38	0.21	−0.39	−0.20	−0.60	−0.40	−0.25
FOREST	−0.61	0.59	−0.77**	0.51	−0.06	0.59	0.20	0.64	0.31	0.01	−0.32	−0.15	0.27	0.14	0.13	0.07	0.47
WOOD	−0.51	0.51	−0.02	−0.36	−0.30	0.51	−0.64	−0.29	0.25	−0.04	−0.08	−0.05	0.13	0.07	0.63	0.32	−0.13
TUNDRA	0.92**	−0.91**	0.59	−0.05	0.32	−0.91**	0.43	−0.22	−0.46	0.03	0.31	0.16	−0.32	−0.17	−0.68	−0.34	−0.23

Note: Significant correlations based on Bonferroni-adjusted probabilities: *, $P \leq 0.05$; **, $P \leq 0.01$.

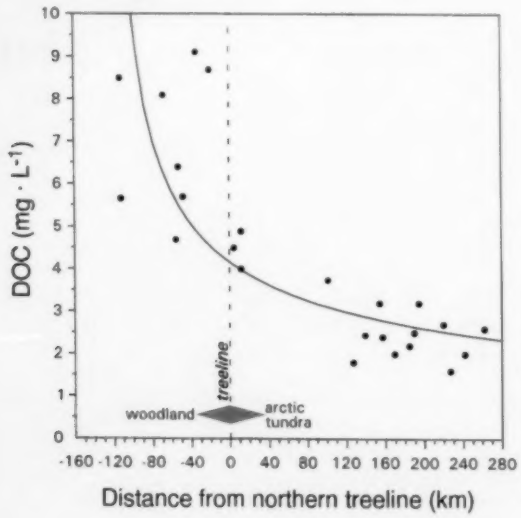


Fig. 4. Plot of DOC concentrations versus distance from the northern treeline in the 24-lake data set.

highest in boreal forest and forest–tundra lakes in the vicinity of Yellowknife (sites 1–3, 20–24) (Table 3). This corresponds to an alkalinity range of approximately 0–1 mequiv. · L^{-1}, which was calculated from [DIC] and pH using the carbonate species relations in Stumm and Morgan (1981).

[DOC] ranged from 1.6 to 9.1 mg · L^{-1}, with a mean of 4.3 mg · L^{-1} for all lakes (Table 3). Because measurements of DOC were usually not included in standard water analyses of lakes in other regions, only a few values were available for comparison. Allochtonous DOC, composed largely of terrestrial humic compounds (humic and fulvic acids), is the primary source of DOC in oligotrophic waters (Wetzel 1983). According to Wetzel (1983) and Engstrom (1987), DOC concentrations in surface waters of small oligotrophic lakes in boreal regions are mainly a function of external humus loading from catchment vegetation and soils. This strong correlation between catchment vegetation and [DOC] is also evident from the Pearson correlation matrix of the present lake set (Table 4), as well as from Fig. 4. Strong colour and high [DOC] were characteristic of shallow basins surrounded by peatlands, especially in the forest–tundra and boreal forest zones.

The three phosphorus variables (TPU, TPF, and SRP) did not exhibit any pronounced gradient such as we recorded, for example, in the Yukon–Tuktoyaktuk survey of lakes (Pienitz et al. 1997). However, levels were generally highest in boreal

© 1997 NRC Canada

Table 4 (*concluded*).

NH_3	TKN	TN	PN	POC	DOC	DIC	SiO_2	Cl	SO_4	Na	K	Ca	Fe	Mn	DIST	FOREST	WOOD
0.35																	
0.29	0.97**																
−0.17	0.02	0.26															
0.24	0.34	0.40	0.30														
0.34	0.89**	0.88**	0.07	0.46													
0.13	0.75*	0.73*	0.03	0.12	0.63												
0.32	0.59	0.56	−0.06	−0.15	0.60	0.69											
−0.03	0.56	0.52	−0.09	−0.01	0.39	0.74*	0.36										
0.11	0.56	0.54	−0.01	0.02	0.47	0.57	0.60	0.54									
0.12	0.77**	0.75*	0.00	0.13	0.61	0.92**	0.55	0.91**	0.64								
0.09	0.76*	0.76**	0.13	0.23	0.61	0.93**	0.52	0.79**	0.64	0.95**							
0.11	0.77**	0.74*	−0.03	0.10	0.67	0.99**	0.72	0.75*	0.60	0.91**	0.91**						
0.24	0.34	0.35	0.09	0.66	0.45	−0.01	−0.15	0.03	0.02	0.12	0.17	0.00					
−0.17	0.16	0.25	0.38	0.42	0.37	0.09	−0.14	0.03	−0.01	0.12	0.23	0.11	0.48				
−0.23	−0.81**	−0.79**	−0.05	−0.31	−0.84**	−0.83**	−0.65	−0.53	−0.46	−0.76*	−0.78**	−0.84**	−0.24	−0.26			
0.18	0.56	0.53	−0.06	−0.12	0.47	0.83**	0.77**	0.70	0.51	0.77**	0.69	0.84**	−0.10	−0.11	−0.62		
0.17	0.39	0.38	0.02	0.46	0.53	0.11	−0.01	−0.14	−0.03	0.08	0.14	0.13	0.35	0.36	−0.51	−0.29	
−0.28	−0.78**	−0.75*	0.03	−0.33	−0.83**	−0.72	−0.57	−0.40	−0.35	−0.65	−0.64	−0.75*	−0.24	−0.25	0.93**	−0.49	−0.70

forest and lowest in arctic tundra lakes. Interestingly, none of the three variables was significantly correlated ($P > 0.05$). TPU concentrations ranged from 3.4 to 12.7 $\mu g \cdot L^{-1}$ (mean = 6.6 $\mu g \cdot L^{-1}$). TN:TP ratios ranged from 18:1 to 71:1, with an average value of 43:1 for all 24 lakes (Table 3).

Concentrations of all nutrients were extremely low, thereby reflecting the oligotrophic nature of these lakes. Similarly low nutrient concentrations have been reported from tundra lakes located northeast of Yellowknife (Moore 1979), where concentrations of TP, NO_3, and SiO_2 usually did not exceed 3.0 $\mu g \cdot L^{-1}$, 10.0 $\mu g \cdot L^{-1}$, and 0.2 $mg \cdot L^{-1}$, respectively. Phosphorus and especially nitrogen are strongly retained by terrestrial tundra ecosystems (Chapin et al. 1980; Dowding et al. 1981), which may explain the tendency for freshwater ecosystems in tundra regions to be limited by both nitrogen and phosphorus. According to the general trophic classification of lakes and reservoirs in relation to phosphorus and nitrogen concentrations (Wetzel 1983), the mean values and ranges of TP and TN concentrations observed in the Yellowknife area indicate that all 24 lakes can be characterized as oligotrophic to ultraoligotrophic.

Iron and manganese

The concentrations of the trace metals Fe and Mn were lower than those reported from the Yukon. Their average concentrations were 46.5 and 4.3 $\mu g \cdot L^{-1}$, respectively (Table 3). Mn concentrations were extremely low (2.0–26.0 $\mu g \cdot L^{-1}$) and decreased below the analytical detection limit of 2.0 $\mu g \cdot L^{-1}$ in most of the lakes. Despite the low concentrations of trace metals, these elements can play an important role as micronutrients in limiting algal photosynthesis in oligotrophic lakes of arctic and temperate regions (e.g., Kalff 1968; Goldman 1972; Wetzel 1983; Hobbie 1984).

Biotic variables

Chlorophyll a

No clear pattern was evident from the CHLAU data, since all lakes had extremely low concentrations. CHLAU values ranged from 0.4 to 1.8 $\mu g \cdot L^{-1}$, averaging 0.9 $\mu g \cdot L^{-1}$ in all lakes (Table 3). Thus, according to Likens' (1975) classification, most of our lakes seemed to be oligotrophic whereas concentrations in two lakes (3, 6) fall into the ultraoligotrophic (0.01–0.5 $\mu g \cdot L^{-1}$) category.

Principal components analysis

The eigenvalues for the four PCA axes were $\lambda_1 = 0.43$, $\lambda_2 = 0.17$, $\lambda_3 = 0.07$, and $\lambda_4 = 0.06$. Thus, the first two axes of the PCA captured 60% of the variance in the data. Almost all variables related to nutrients and major ions had high scores

© 1997 NRC Canada

on the major PCA axis 1, which explained almost 43% of the total variance. The contrast in the relative size of the first two axes suggests that there is one overriding gradient of variation in the chemical data, namely axis 1 with the gradient from high to low organically bound nutrients and major ions. The passive variable LAT was negatively correlated with TEMP, COND, nutrients (TN, TKN, TPU, DIC, DOC, and SiO_2), and major ions (K, Na, Ca, SO_4, and Cl), as indicated by the arrows pointing more or less into the opposite direction of the LAT arrow (Fig. 2). Thus, the first axis effectively contrasted boreal forest and forest–tundra lakes (sites 1–4, 22–24), having relatively high nutrient and major ion levels, with dilute arctic tundra sites (6–18), which clustered in the lower two quadrants of the ordination biplot. Site 21, which scored highest on axis 2, was a strongly coloured, shallow lake, which had the highest Fe and POC concentrations, as well as the lowest water transparency.

In a PCA correlation biplot, variables with high positive correlations generally have small angles between their biplot arrows, identifying groups of mutually redundant variables, such as TN and TKN, as well as K, Na, and Ca (Fig. 2). Variables with long arrows have high variance and are often the most important within the data (Jongman et al. 1987). The direction of each arrow indicates ascending values for each variable.

The lack of statistically significant relationships between CHLAU and the organically bound nutrients was no surprise, as chlorophyll *a* concentration largely depends on the loading of inorganic nutrients, which was not measured in these oligotrophic systems.

Fe, POC, TRANS, and DEPTH are loading highly on the second PCA axis, with Fe and POC being negatively correlated with TRANS and DEPTH. Their distributions basically illustrate a gradient in water colour, with lowest transparencies occurring in shallow lakes characterized by high concentrations in POC and Fe.

As expected, the passive variable LAT, which scored highly on the first PCA axis, exhibited highly significant negative correlations with both TEMP ($P < 0.01$) and DOC ($P < 0.05$) (Table 4; Fig. 2), thereby reflecting the strong climatic and vegetational gradients of the study area. Because the lakes were visited along a northeast-trending sampling transect (i.e., at a right angle to the northern treeline), LAT and LONG were highly negatively correlated ($r = -20.96$, $P < 0.01$) (Table 4).

Conclusions

The physical and chemical data gathered for the 24 study lakes reveal comparatively homogeneous conditions, with consistently low nutrient and ion concentrations and relatively little variability among sites. The dilute nature of the study lakes is typical for Shield regions, as the lakes drain bedrock that is highly resistant to chemical weathering. Our results also confirm that, as has been determined elsewhere, watersheds with predominantly granitic parent materials possess lakes with low surface water alkalinities that are potentially sensitive to the effects of acid deposition.

Inputs of materials from terrestrial ecosystems strongly regulate the functioning of arctic lakes (Kling 1995) and are controlled to a great extent by vegetation that influences the composition and fluxes of elements in watersheds (Likens et al. 1977; Cornwell 1992). This is partly related to the storage and release of ions in living and dead organic matter and enhanced weathering owing to plant-derived organic acids. Plants also influence the thaw depth of arctic tundra soils, with unvegetated sites generally having a greater active zone (Brown and Berg 1980).

The concentration of DOC seemed to be strongly related to catchment vegetation (Table 4; Fig. 2). The discrepancy between DOC concentrations of lakes in forested and unforested watersheds may be attributed largely to differences in the production and sequestering of humic compounds in the vegetation and soils of the drainage basin (e.g., Engstrom 1987; Driscoll et al. 1987). Although humus loading to surface waters from sparsely vegetated woodlands is generally lower than that from forested catchments (Engstrom 1987), the elevated concentrations of DOC were most apparent in forest–tundra sites, where most lakes were tea-coloured and exhibited the lowest water transparencies. The latter may be explained by the shallowness of these lakes as well as inhibition of downward percolation of humic materials to deeper soil horizons owing to the presence of more extensive permafrost in the area.

The significant differences in ionic concentrations observed between lakes in the Yellowknife area and those along the Yukon–Tuktoyaktuk transect most likely reflect the differences between freshwater systems located on different types of bedrock. Ionic concentrations in lakes depend largely on the soil characteristics and, ultimately, on the parent bedrock of the drainage basin (Hobbie 1984).

Besides bedrock geology, vegetation type, and climate, surface water chemistry may also be influenced by the age of glacial drift in a drainage basin and thus the extent of primary rock exposure, soil development, and weathering (Kling et al. 1992). Lakes lying in younger glacial drift usually have higher conductivities and are strongly enriched in major ions when compared with lakes surrounded by older and more weathered drift (Kling et al. 1992). This relationship between lake water chemistry and the age and amount of unconsolidated sediments covering the land surface seems to be an important factor in determining nutrient availability and ionic concentrations in our study sites. Likewise, soil nutrient deficiencies imposed upon terrestrial vegetation by the shallow, sandy brunisols of the Canadian Shield have been reported by Larsen (1989) and Timoney et al. (1992).

In general, the narrow ranges in almost all of the chemical and physical variables indicate that limnological conditions were relatively homogeneous as compared with those reported from Yukon lakes (Pienitz et al. 1997).

Acknowledgements

This study was financed by the Natural Sciences and Engineering Research Council of Canada via a grant to J.P.S., a Canadian Government Scholarship to R.P., and Green Plan support on aquatic impacts of ultraviolet light to D.R.S.L. We thank Dr. B. Zeeb for help with the field work, which was supplemented by a grant from the Northern Studies Training Program.

References

Brown, J., and Berg, R.L. 1980. Environmental engineering and ecological baseline investigations along the Yukon River – Prudhoe

© 1997 NRC Canada

Bay Haul Road. CRREL report No. 80–19. U.S. Army Cold Regions Research and Engineering Laboratory, Hanover, N.H.

Bryson, R.A. 1966. Air masses, streamlines, and the boreal forest. Geogr. Bull. **8**: 228–269.

Chapin, F.S., III, Miller, P.C., Billings, W.D., and Coyne, P.I. 1980. Carbon and nutrient budgets and their control in coastal tundra. *In* An arctic ecosystem: the coastal tundra at Barrow. *Edited by* J. Brown, P.C. Miller, L.L. Tieszen, and F.L. Bunnell. Dowden, Hutchinson & Ross, Inc., Stroudsburg, Pa. pp. 458–482.

Cornwell, J.C. 1992. Cation export from Alaskan arctic watersheds. Hydrobiologia, **240**: 15–22.

Cunningham, M.P., and Lambert, R.S.J. 1989. Petrochemistry of the Yellowknife volcanic suite at Yellowknife, N.W.T. Can. J. Earth Sci. **26**: 1630–1646.

Dowding, P., Chapin, F.S., III, Weilgolaski, F.E., and Kilfeather, P. 1981. Nutrients in tundra ecosystems. *In* Tundra ecosystems, a comparative analysis. *Edited by* L.C. Bliss, O.W. Heal, and J.J. Moore. Cambridge University Press, Cambridge. pp. 647–683.

Driscoll, C.T., Yatsko, C.P., and Unangst, F.J. 1987. Longitudinal and temporal trends in the water chemistry of the North Branch of the Moose River. Biogeochemistry, **3**: 37–62.

Ecoregions Working Group. 1989. Ecoclimatic regions of Canada, first approximation. Ecological land classification series No. 23. Ecoregions Working Group, Environment Canada, Ottawa, Ont.

Engstrom, D.R. 1987. Influence of vegetation and hydrology on the humus budgets of Labrador lakes. Can. J. Fish. Aquat. Sci. **44**: 1306–1314.

Environment Canada. 1979. Analytical methods manual. Inland Waters Directorate, Water Quality Branch, Ottawa, Ont.

Environment Canada. 1985–1991. Yellowknife ice freeze-up and ice break-up reports. Atmospheric Environment Service, Yellowknife, N.W.T.

Falk, M.R. 1979*a*. Biological and limnological data on ten lakes surveyed in the Northwest Territories, 1971–1972. Fisheries and marine service data report No. 129. Department of Fisheries and the Environment, Winnipeg, Man.

Falk, M.R. 1979*b*. Biological and limnological data on lakes along the Ingraham Trail, Northwest Territories, 1973–1974. Fisheries and marine service data report No. 125. Department of Fisheries and the Environment, Winnipeg, Man.

Frith, R.A. 1987. Precambrian geology of the Hackett River area, District of Mackenzie, N.W.T. Mem. Geol. Surv. Can. No. 417.

Goldman, C.R. 1972. The role of minor nutrients in limiting the productivity of aquatic ecosystems. Limnol. Oceanogr. (Spec. Symp.) **1**: 21–33.

Hare, F.K., and Ritchie, J.C. 1972. The boreal bioclimates. Geogr. Rev. **62**: 334–365.

Healey, M.C., and Woodall, W.L. 1973*a*. Experimental cropping of lakes. II. Physical and chemical features of the lakes. Fish. Res. Board Can. Tech. Rep. No. 384.

Healey, M.C., and Woodall, W.L. 1973*b*. Limnological surveys of seven lakes near Yellowknife, Northwest Territories. Fish. Res. Board Can. Tech. Rep. No. 407.

Henderson, J.B. 1970. Stratigraphy of the Yellowknife Supergroup, Yellowknife Bay – Prosperous Lake area, District of Mackenzie. Geol. Surv. Can. Pap. No. 70-26.

Henderson, J.B. 1981. Archean basin evolution in the Slave Province, Canada. *In* Precambrian plate tectonics. *Edited by* A. Kröner. Elsevier Science Publishers B.V., Amsterdam, The Netherlands. pp. 213–235.

Hobbie, J.E. 1984. Polar limnology. *In* Ecosystems of the World 23: lakes and reservoirs. *Edited by* F.B. Taub. Elsevier Science Publishers B.V., Amsterdam, The Netherlands. pp. 63–105.

Hobbie, J.E., Deegan, L.A., Peterson, B.J., Rastetter, E.B., Shaver, G.R., Kling, G.W., O'Brien, W.J., Chapin, F.S.T., Miller, M.C., Kipphut, G.W., Bowden, W.B., Hershey, A.E., and McDonald, M.E. 1995. *In* Long-term measurements at the arctic LTER site. Ecological time series. *Edited by* T.M. Powell and J.H. Steele. Chapman and Hall, New York. pp. 391–409.

Jongman, R.H.G., Ter Braak, C.J.F., and Van Tongeren, O.F.R. 1987. Data analysis in community and landscape ecology. Pudoc, Wageningen, The Netherlands.

Kalff, J. 1968. Some physical and chemical characteristics of arctic fresh waters in Alaska and northwestern Canada. J. Fish. Res. Board Can. **25**: 2575–2587.

Kling, G.W. 1995. Land–water interactions: the influence of terrestrial diversity on aquatic ecosystems. *In* Arctic and alpine biodiversity: patterns, causes and ecosystem consequences. Ecological studies No. 113. *Edited by* F.S. Chapin III and C. Körner. Springer-Verlag, New York. pp. 297–310.

Kling, G.W., O'Brien, W.J., Miller, M.C., and Hershey, A.E. 1992. The biogeochemistry and zoogeography of lakes and rivers in arctic Alaska. Hydrobiologia, **240**: 1–14.

Larsen, J.A. 1989. The northern forest border in Canada and Alaska. Ecological Studies No. 70. Springer-Verlag, New York.

Likens, G.E. 1975. Primary production of inland aquatic ecosystems. *In* The primary productivity of the biosphere. *Edited by* H. Lieth and R.W. Whittaker. Springer-Verlag, New York. pp. 185–202.

Likens, G.E., Bormann, F.H., Pierce, R.S., Eaton, J.S., and Johnson, N.M. 1977. Biogeochemistry of a forested ecosystem. Springer-Verlag, New York.

Livingstone, D.A. 1966. Alaska, Yukon, Northwest Territories, and Greenland. *In* Limnology in North America. *Edited by* D.G. Frey. University of Wisconsin Press, Madison, Wis. pp. 559–574.

Moore, J.W. 1979. Distribution and abundance of attached, littoral algae in 21 lakes and streams in the Northwest Territories. Can. J. Bot. **57**: 568–577.

Moore, J.W. 1981. Benthic algae in littoral and profundal areas of a deep subarctic lake. Can. J. Bot. **59**: 1026–1033.

Moore, J.W., Sutherland, D.J., and Beaubien, V.A. 1979. Algal and invertebrate communities in three subarctic lakes receiving mine wastes. Water Resour. Bull. **13**: 1193–1202.

Padgham, W.A., and Fyson, W.K. 1992. The Slave Province: a distinct Archean craton. Can. J. Earth Sci. **29**: 2072–2086.

Patalas, K. 1984. Mid-summer mixing depths of lakes of different latitudes. Verh. Int. Ver. Theor. Angew. Limnol. **22**: 97–102.

Pienitz, R. 1993. Paleoclimate proxy data inferred from freshwater diatoms from the Yukon and the Northwest Territories. Ph.D. thesis, Queen's University, Kingston, Ont.

Pienitz, R., and Smol, J.P. 1993. Diatom assemblages and their relationship to environmental variables in lakes from the boreal forest – tundra ecotone near Yellowknife, Northwest Territories, Canada. Hydrobiologia, **269–270**: 391–404.

Pienitz, R., Smol, J.P., and Lean, D.R.S. 1997. Physical and chemical limnology of 59 lakes located between the southern Yukon and the Tuktoyaktuk Peninsula, Northwest Territories (Canada). Can. J. Fish. Aquat. Sci. **54**: 330–346.

Rawson, D.S. 1960. A limnological comparison of twelve large lakes in northern Saskatchewan. Limnol. Oceanogr. **5**: 195–211.

Robertson, D.M. 1989. The use of lake water temperature and ice cover as climatic indicators. Ph.D. thesis, University of Wisconsin-Madison, Madison, Wis.

Rowe, J.S. 1972. Forest regions of Canada. Canadian Forest Service, Ottawa, Ont.

Shuter, B.J., Schlesinger, D.A., and Zimmerman, A.P. 1983. Empirical predictors of annual surface water temperature cycles in North American lakes. Can. J. Fish. Aquat. Sci. **40**: 1838–1845.

Stumm, W., and Morgan, J.J. 1981. Aquatic chemistry. 2nd ed. John Wiley & Sons, New York.

Ter Braak, C.J.F. 1990*a*. CANOCO: a FORTRAN program for CANOnical Community Ordination. Microcomputer Power, Ithaca, N.Y.

© 1997 NRC Canada

Ter Braak, C.J.F. 1990*b*. Update notes: CANOCO version 3.10. Agricultural Mathematics Group, Wageningen, The Netherlands.

Tibbatts, W.L., Gregor, D.J., and James, L. 1987. Aquatic sensitivity mapping for acid deposition: Northwest Territories, Canada. Environment Canada, Inland Waters Directorate – Western and Northern Region, Regina, Sask.

Timoney, K.P., La Roi, G.H., Zoltai, S.C., and Robinson, A.L. 1992. The high subarctic forest–tundra of northwestern Canada: position, width, and vegetation gradients in relation to climate. Arctic, **45**: 1–9.

Van Tongeren, O.F.R., Van Liere, L., Gulati, R.D., Postema, G., and Boesewinkel-De Bruyn, P.J. 1992. Multivariate analysis of the plankton communities in the Loosdrecht lakes: relationship with the chemical and physical environment. Hydrobiologia, **233**: 105–117.

Wallace, R.R., and Hardin, M.J. 1975. Chemical and biological characteristics of seepages from tailings areas at Giant Yellowknife Mines Ltd., into Great Slave Lake, Northwest Territories, in 1974. Report No. 5-NW-75-1. Canadian Department of Fisheries and Environment, Environmental Protection Service (Northwest Region), Yellowknife, N.W.T.

Welch, H.E. 1985. Introduction to limnological research at Saqvaqjuac, northern Hudson Bay. Can. J. Fish. Aquat. Sci. **42**: 494–505.

Welch, H.E., and Legault, J.A. 1986. Precipitation chemistry and chemical limnology of fertilized and natural lakes at Saqvaqjuac, N.W.T. Can. J. Fish. Aquat. Sci. **43**: 1104–1134.

Wetzel, R.G. 1983. Limnology. 2nd ed. Saunders College Publishing, Philadelphia, Pa.

Wilkinson, L. 1988. SYSTAT: the system for statistics. SYSTAT Inc., Evanston, Ill.

Zar, J.H. 1984. Biostatistical analysis. 2nd ed. Prentice-Hall Inc., Englewood Cliffs, N.J.

© 1997 NRC Canada

Factors affecting the production of zooplankton in lakes[1]

B.J. Shuter and K.K. Ing

Abstract: Multiple regression analysis and analysis of covariance were used to assess the degree to which observed variation in rates of production among 108 lake zooplankton populations could be accounted for by variation in physical and biological factors. Seventy-six percent of the observed variation in growing season weight-specific production rates could be accounted for by differences in temperature, length of growing season, and taxonomic group (Rotifera, Chydoroidea, Cyclopoida, Calanoida, ordered from highest rate to lowest). Within taxonomic groups, effects of population biomass density, individual body size, and food availability were not detected. Fifty-eight percent of the observed variation in growing season population biomass density could be accounted for by differences in individual body size, mean chlorophyll concentration, and lake mean depth. Twenty-four percent of the observed variation in growing season length could be accounted for by differences in mean chlorophyll concentration and taxonomic group. Our findings suggest a simple model of the seasonal production cycle for limnetic zooplankton in which weight-specific rates of biomass production are largely set by temperature, and levels of biomass accumulation are largely set by food resource availability and individual body size. We briefly discuss the implications of this model for predicting the effects of climate change on lake productivity.

Résumé : L'analyse de régression multiple et l'analyse de covariance ont été utilisées pour évaluer dans quelle mesure la variation observée dans les taux de production de 108 populations de zooplancton de lac pouvait s'expliquer par la variation de facteurs physiques et biologiques. Soixante-seize pour cent de la variation observée dans les taux de production spécifiques du poids au cours de la saison de végétation pouvait s'expliquer par des différences touchant la température, la longueur de la saison de végétation et le groupe taxinomique (rotifères, chydoroïdés, cyclopoïdés, calanoïdés, par ordre décroissant du taux de production). Au sein des groupes taxinomiques, on n'a pas décelé d'effets liés à la densité de biomasse de la population, à la taille corporelle individuelle ni à la disponibilité des aliments. Cinquante-huit pour cent de la variation observée dans la densité de biomasse de la population au cours de la saison de végétation pouvait s'expliquer par des différences touchant la taille corporelle individuelle, la concentration moyenne de chlorophylle et la profondeur moyenne des lacs. Vingt-quatre pour cent de la variation observée dans la longueur de la saison de végétation pouvait s'expliquer par les différences touchant la concentration moyenne de chlorophylle et le groupe taxinomique. Nos constatations révèlent un modèle simple du cycle de production saisonnier pour le zooplancton limnétique, dans lequel les taux de production de biomasse spécifiques du poids sont en grande partie déterminés par la température, alors que les concentrations de biomasse sont en grande partie déterminées par la disponibilité des aliments et la taille corporelle individuelle. Nous discutons brièvement des implications de ce modèle pour la prévision des effets des changements climatiques sur la productivité des lacs.
[Traduit par la Rédaction]

Introduction

Empirical studies have demonstrated that biomass production in natural animal populations varies systematically with population biomass, body size, and broad taxonomic group (Humphreys 1979, 1981; Banse and Mosher 1980; Dickie et al. 1987). A series of recent studies on aquatic invertebrates (freshwater limnetic, Plante and Downing 1989; freshwater lotic, Morin and Bourassa 1992; marine benthic, Tumbiolo and Downing 1994; marine copepods, Huntley and Lopez 1992) has shown that much of the observed variation in annual production by natural populations of these organisms can be accounted for by variations in environmental temperature, population biomass density, and body size. Our objective in this paper is to examine the effects of these, and other variables, on the production of natural populations of limnetic zooplankton.

Most empirical studies of production have focused on observed variation in annual rates. However, the literature on limnetic zooplankton contains considerable information on seasonal variation in daily production rates within a population, as well as information on variation in annual rates between populations. We wished to examine both types of data within a consistent theoretical framework. We used the recent findings of Huntley and Lopez (1992) to develop such a

Received June 21, 1995. Accepted August 9, 1996.
J12967

B.J. Shuter and K.K. Ing. Harkness Laboratory of Fisheries Research and Aquatic Ecosystem Science Section, Ontario Ministry of Natural Resources, Box 5000, Maple, ON L6A 1S9, Canada.

[1] Contribution No. 95-07, Aquatic Ecosystem Science Section, Ontario Ministry of Natural Resources.

© 1997 NRC Canada

framework. These authors examined variation in daily weight-specific production rates of marine copepods, in both laboratory and field situations, and found that most of the observed variation was linked to variations in water temperature, according to a simple Van't Hoff relationship. Let us assume that a similar relationship holds for freshwater zooplankton:

$$(1) \quad [P/B]_{daily} = a \cdot e^{b \cdot T_{daily}}$$

where $[P/B]_{daily}$ is the daily weight-specific rate of production and T_{daily} is the daily water temperature. Given this, then annual production (P_{annual}) can be written as

$$(2) \quad P_{annual} \approx [\overline{P/B}]_{daily} \cdot \overline{B}_{gs} \cdot gs$$

where $[\overline{P/B}]_{daily}$ is the average daily weight-specific rate of production over the growing season, $\overline{B}_{gs}$ is the average population biomass over the growing season, and gs is the length of the growing season.

If the temperature covers less than a 15°C range over the growing season and b is about 0.045 (range of Huntley and Lopez (1992) estimates for b: 0.04–0.05), then it is easy to show numerically that the following equation holds:

$$(3) \quad [\overline{P/B}]_{daily} \approx a \cdot e^{b \cdot T_{gs}}$$

where T_{gs} is the average water temperature over the growing season. Substituting this into equation 2 yields

$$(4) \quad P_{annual} \approx a \cdot e^{b \cdot T_{gs}} \cdot \overline{B}_{gs} \cdot gs$$

and we then rearrange terms to arrive at

$$(5) \quad P_{annual} / \overline{B}_{gs} \approx a \cdot e^{b \cdot T_{gs}} \cdot gs$$

The relationship between daily, weight-specific production rates and temperature can be recovered from information on annual production and growing season averages for both biomass and water temperature (eq. 5). Annual production is itself the product of three quite distinct variables (eq. 2): the average daily weight-specific production rate, the average biomass over the growing season, and the length of the growing season. We will analyze observed variation in each of these quantities for a wide variety of natural zooplankton populations. Our data set is derived from studies on 38 lakes that span the globe (Tables 1 and 2), from the High Arctic (74°N) through tropical Africa to New Zealand (44°S).

Methods

Data acquisition and standardization

As is common in comparative studies, many different workers were involved in generating the data for our analyses. In general, we have assumed that those workers had the experience and judgment necessary to collect useable data from their specific systems and to estimate population production from that data. Studies were checked in several ways to help buttress this assumption.

(*i*) Studies were included only if they were published in the primary scientific literature and had, as a major objective, estimation of population production.

(*ii*) Typically, these studies met the mesh size guidelines for sampling gear set out by De Bernardi (1984): 43% used mesh sizes ≤50 μm, while 49% used mesh sizes in the range >50–126 μm; larger mesh sizes were used for the larger species (i.e., daphnids and copepods); De Bernardi (1984) recommended 126 μm for larger zooplankton and 50 μm for the smaller species.

(*iii*) Studies were included only if the method used to calculate production met the guidelines set by Rigler and Downing (1984): (*a*) production estimates derived by assuming equality of the turnover rates for population numbers and population biomass were accepted for organisms with simple life cycles (i.e., rotifers) without additional confirmation; for cladocerans, they were accepted when they could be checked against other data provided in the original paper; for copepods and cyclopoids, they were rejected; (*b*) production estimates based on reference values for the *P/B* ratio were rejected; (*c*) production estimates based on values for the durations of different life stages were used only if durations were estimated directly in the field, or were estimated from laboratory data, using observed field temperatures.

(*iv*) Studies were included only if production values from all life stages, including egg production by adults, were part of the population production estimates.

We began with the list of studies used by Plante and Downing (1989) and then extended it by systematically searching the more recent (1988–1994) literature for papers reporting production rates of limnetic zooplankton. The following data were extracted from these sources (Table 1): (*i*) daily estimates for biomass, production rate, and water temperature for at least one growing season (daily data set); (*ii*) annual estimates of total production, average biomass, and average water temperature (annual data set); (*iii*) within at least one annual cycle, estimates of growing season length, average biomass over the growing season, and average water temperature over the growing season (growing season data set); and (*iv*) measures of zooplankton food resource availability (dissolved phosphorus and chlorophyll concentrations), lake mean depth, pH, and other environmental variables.

All production and abundance (numbers and biomass) estimates were standardized to a common spatial unit: 1 m^2 of lake surface area. All production and biomass estimates were standardized to a common mass unit: grams dry weight. When unit conversion was required, appropriate conversion factors were taken from the original study whenever possible. If no conversion information was provided in the original study, the following standard values were used: dry weight = carbon/0.44 (Burgis 1971; Green 1976; Gophen 1978; Herzig et al. 1980; Andrew 1983; Mason and Abdul-Hussein 1991; Huntley and Lopez 1992) and species-specific and congener-specific conversions from calories to dry weight, as listed in Cummins and Wuycheck (1971).

In studies of multiple years or stations, atypical years or stations, as indicated by the author(s), were excluded from our data base. Otherwise, averages for all years or stations were used.

With the exception of two unusual populations from one lake (Lake Tjeukemeer), no attempts were made to recalculate production estimates from available data. In the two cyclopoid populations from Lake Tjeukemeer, weight-specific production values reported for nauplii far exceeded (by over 400%) values exhibited by the older life stages in these populations and by the other cyclopoid populations in our data base. Because of these anomalously high values, we recalculated population production for each population, assuming that the weight-specific production rate of naupliar stages was identical with that of the older life stages in the population. These adjusted values were used throughout our analyses.

Defining annual estimates and growing season estimates

There are inconsistencies in the literature regarding definitions for annual estimates of production and biomass and growing season estimates of production and biomass (Edmondson 1974). Some authors equate growing season estimates with annual estimates, and some do

© 1997 NRC Canada

Table 1. Summary table of zooplankton populations by lake and species group.

Continent[a]	Lake	Species[b]	Data type	Reference(s)
1	Char Lake	5, 7	A, G	Rigler et al. 1974; MacCallum 1971
1	Clear Lake	2, 5	D	Schindler 1972
1	Lake Ontario	3, 3, 3, 3, 6, 6	A, G	Borgmann et al. 1984
1	Mirror Lake	2, 4, 7	A, G	Makarewicz and Likens 1979
1	Pyramid Lake	5	A, G	Herzig et al. 1980
2	Broa Reservoir	5	A, G	Matsumura-Tundisi et al. 1989; Rocha and Matsumura-Tundisi 1984
3	Ardleigh Reservoir	3, 3	A, D, G	Mason and Abdul-Hussein 1991
3	Eglwys Nyndd Reservoir	6	A, G	George 1976
3	Farmoor I Reservoir	3	A, D, G	Jones et al. 1979
3	Lake Akulkino	1, 7, 7	A, G	Winberg et al. 1973
3	Lake Constance	3, 3	A, G	Geller 1989; Sommer 1985; Trippel 1991
3	Lake Esrom	5, 7	A, D, G	Bosselman 1975, 1979*a*, 1979*b*
3	Lake Krivoe	2, 6, 7, 7, 4	A, G	Alimov et al. 1972
3	Lake Krugloe	2, 2, 4, 6, 7	A, G	Alimov et al. 1972
3	Lake Kvernavatnet	3	A, D, G	Borsheim et al. 1988
3	Lake Myvatn	3, 6, 7, 7, 7	A, G	Adalsteinsson 1979
3	Lake Naroch	2, 6	A, D, G	Babitskiy 1970, 1988
3	Lake Ormajarvi	7, 7, 7, 7	A, D	Cajander 1983
3	Lake Port-Bielh	3, 5	A, D, G	Rey and Capblancq 1975
3	Lake Taltowisko	2, 5, 6, 7	A, G	Hillbricht-Ilkowska and Weglenska 1970
3	Lake Zelenetzkoye	1, 7, 7	A, G	Winberg et al. 1973
3	Lough Neagh	7, 7, 7, 7, 7	A, D, G	Andrew and Fitzsimons 1992
3	Mikolajskie Lake	2, 5, 6, 7	A, G	Hillbricht-Ilkowska and Weglenska 1970
3	Ovre Heimdalsvatn	2, 3, 6	A, G	Larsson 1978
3	Queen Mary Reservoir	3	A, D, G	Andrew 1983
3	Red Lake	2, 2, 3, 3, 3, 5, 6, 6, 7	A, G	Andronikova et al. 1970
3	Tjeukemeer	3, 3, 6, 6	A, D, G	Vijverberg and Richter 1982*a*, 1982*b*; Vijverberg and Frank 1976
4	Lake Kasumigaura	2, 3, 3, 3	A, D, G	Hanazato and Yasuno 1985
5	Lake Awasa	2, 6, 6	A, G	Mengestou and Fernado 1991*a*, 1991*b*
5	Lake Chad	2, 2, 3, 3, 3, 3, 5, 6	A, D, G	Lévêque and Saint-Jean 1983
5	Lake George	6	A, G	Burgis and Walker 1972; Burgis 1971, 1974
5	Lake Kinneret	2, 4, 6	A, D, G	Gophen 1978, 1988; Gophen et al. 1990
5	Lake Naivasha	5, 6	A, G	Mavuti 1990, 1994
5	Lake Nakuru	5, 7, 7	A, G	Vareschi 1982; Vareschi and Jacobs 1984, 1985; Vareschi and Vareschi 1984
6	Lake Hayes	5	A, D, G	Burns 1979
6	Lake Okaro	3, 3, 5	A, D, G	Forsyth and James 1991
6	Lake Ototoa	5	A, D, G	Green 1976
6	Lake Samsonvale	3, 5	A, D, G	King and Greenwood 1992*a*, 1992*b*

Note: Types of data obtained from each population are also listed: A, annual production; D, daily production; G, growing season.
[a]1, North America; 2, South America; 3, Europe; 4, Asia; 5, Africa; 6, Australia.
[b]1, Crustacea; 2, Cladocera; 3, Chydoroidea; 4, Copepoda; 5, Calanoida; 6, Cyclopoida; 7, Rotifera.

not. Such inconsistencies can produce large and spurious differences in average biomass estimates, particularly for temperate zooplankton populations. Such populations typically have a short growing season, when biomass and production are at detectable levels, and a long period of dormancy, when biomass and production are often undetectable. Under these circumstances, annual production (P) and growing season production are essentially identical, but annual average biomass ($\overline{B}_{ann}$) is effectively equal to the growing season average biomass ($\overline{B}_{gs}$) multiplied by the fraction of the year taken up by the growing season. When the growing season is short, $\overline{B}_{ann}$ will be low and will not reflect the average amount of biomass responsible for generating the production observed over the year.

Plante and Downing (1989) based their analyses on annual averages for both population biomass and water temperature, and, for

© 1997 NRC Canada

comparative purposes, we will use this approach on some of our data sets. However, we will focus our analyses around eq. 5 because it deals explicitly with the growing season effect.

Only a minority of authors actually estimated growing season length (gs) and each used his own, often undefined, protocol. We wished to develop consistent estimates of growing season length for as many populations as possible. Because considerable variation existed among populations in the amount of data available for estimating gs, we developed three similar definitions (each with different data demands) to maximize the number of populations for which we could obtain roughly consistent estimates of gs. In order of preference, the three definitions are as follows: (*i*) that part of a year for which monthly biomass values exceeded 20% of the peak monthly biomass value for the year (54 of our 105 gs estimates were obtained using this protocol), (*ii*) that part of a year for which monthly production values exceeded 20% of the peak monthly production value for the year (5 of 105 estimates), and (*iii*) that part of the year identified by source authors as the "vegetative" or "growing" season (46 of 105 estimates). With the growing season defined, values for gs, $\overline{B}_{gs}$, and $\overline{B}_{ann}$ were readily determined. Because most production occurs during the growing season, we assumed a negligible difference between P and growing season production and equated the two.

Other variables

Estimates for annual (T_{ann}) and growing season (T_{gs}) average water temperatures were derived from temperatures recorded in the upper isothermal layer of each lake. Arctic and tropical lakes were essentially isothermal from top to bottom throughout the year; hence, T_{ann} and T_{gs} equated to whole-lake average temperatures. For temperate-zone dimictic lakes, T_{ann} and T_{gs} were derived from mixed-layer temperatures during the ice-free period and whole-lake temperatures during the ice-cover period (i.e., all lakes were essentially isothermal from top to bottom during the ice-cover period). In dimictic lakes, some zooplankton populations almost certainly spend time in both epilimnion and hypolimnion. Thus, our measure of temperature must be regarded as merely an index of the average temperature experienced by these populations. Although this introduces some unavoidable noise into our data set, we feel that its overall influence on our results will be minimal because (*i*) the only strict cold-water form in our data set (*Limnocalanus macrurus*) is from an essentially isothermal arctic lake, and therefore, our protocols will provide an accurate measure of its temperature exposure; (*ii*) in our data set, all species from dimictic lakes spend all or part of their time in the epilimnion; in one detailed study (Geller 1989), the time budgets of migratory and strictly epilimnetic species were examined and the difference in average temperature exposure between the two species was small (3–5°C) relative to the overall temperature range in the data set (about 28°C); and (*iii*) observations at the extremes of temperature exposure (arctic and tropical lakes) will be most influential in defining the character of whatever temperature effect exists in the data set; all of these estimates are from essentially isothermal lakes and therefore are not affected by the vertical migration problem.

We used the following protocols for estimating water temperatures. When intraannual time series of lake water temperature estimates were available, mean annual water temperature and mean water temperature during the growing season were calculated from these estimates. Mean annual air temperature (air) for the lake was obtained from the source reference, or was estimated from a world climatic summary as needed (Wernstedt 1972). When direct estimates of lake water temperatures were not available, estimates for T_{ann} were obtained from the mean annual air temperature and mean lake depth, using a multiple regression equation derived from those lakes with direct estimates of T_{ann}, air, and mean lake depth ($R^2 = 0.97$, $N = 37$, $p < 0.001$). This equation was used to derive T_{ann} estimates for 12 of 108 populations (2 of 38 lakes). When direct estimates of growing season water temperatures were not available, estimates for T_{gs} were obtained from T_{ann} and gs estimates, using a multiple regression equation derived from those populations with direct estimates of T_{gs}, T_{ann}, and gs ($R^2 = 0.90$, $N = 77$, $p < 0.001$). This equation was used to derive T_{gs} estimates for 23 of 87 populations (from 9 of 38 lakes).

In addition to temperature, we examined the relative importance of the following variables in predicting annual production: (*i*) biotic variables: taxonomic group, average annual population biomass ($\overline{B}_{ann}$), average growing season population biomass ($\overline{B}_{gs}$), adult body mass (M_{ad}), and average individual mass in the population (M_{av}); (*ii*) climatic and morphometric variables: maximum solar radiation (mxsol) and lake mean depth (Z); and (*iii*) indices of zooplankton food resource availability: mean annual dissolved phosphorus concentration (phos) and mean chlorophyll concentration (chl) over the vegetative season (i.e., usually equivalent to the ice-free period).

We used two criteria to select variables: first, previous recognition in the literature (e.g., Brylinsky 1980; Straskraba 1980; Plante and Downing 1989) that the variable was associated with invertebrate productivity, and second, availability of estimates for a substantial (>20) number of populations. Protocols for estimating variables are outlined below.

We used two quite different measures of body size: M_{ad} reflects the ultimate size attainable by individuals in a population, while M_{av} reflects the age and size structure of the population over the growing season.

Estimates of adult individual size (M_{ad}) were standardized to mean female adult mass (milligrams dry weight), because we found that estimates of mean female mass were more readily available in the original articles reporting production. When such direct estimates were not available, we derived indirect estimates using one of the following sources (in order of preference): (*i*) different source article describing the same population, (*ii*) species-specific size data provided in summary publications (Edmondson 1959; Clegg 1963; Balcer et al. 1984; McCauley 1984; Culver et al. 1985; Pennak 1989; Thorp and Covich 1991), (*iii*) species-specific scale diagrams in summary publications, and (*iv*) average adult mass for the appropriate genus, as reported in the source publication providing production and biomass estimates or as calculated from our accumulated data sets. Where necessary, length estimates were converted to mass estimates using the appropriate species-specific length–weight conversion equation as found in the original source reference or in summary publications as described above.

Following Morin and Bourassa (1992), we estimated the average mass of an individual in the population (M_{av}) by dividing the mean annual biomass by the mean annual numerical abundance. We estimated mxsol from latitude using fig. 3.2 in Straskraba (1980). Values for our measures of food resource availability (phos, chl) were obtained from the reference providing zooplankton production data, or other published papers dealing with the same lake. For lakes where both measures are available ($N = 27$), the relationship between chl and phos is quantitatively similar to the chlorophyll–phosphorus relationships reported for other lake data sets by Pace (1984), Mazumder (1994), and Chow-Fraser et al. (1994).

Units, symbols, and summary statistics for all variables are provided in Table 2.

Statistical analysis

A tacit assumption built in to equations 2–5 is that dividing population production by population biomass effectively removes the influence of biomass on production. Downing (1984) has argued that this assumption is untenable because of the inevitable reductions in population production at high levels of population biomass as a result of density-dependent restrictions on population growth. He suggested (Downing 1984; Plante and Downing 1989) that differences in total population production rates should be examined directly using regression techniques and the effects of population biomass should be removed empirically by including population biomass as an independent variable in the analysis.

© 1997 NRC Canada

Table 2. Summary statistics for environmental characteristics of all lakes in the study (maximum $N = 38$) and characteristics of zooplankton populations included in the annual data set ($N = 108$) and growing season data set ($N = 87$).

	Environmental characteristics of lakes					
	Symbol	N (lakes)	Minimum	Mean	Maximum	Standard deviation
Maximum solar radiation ($J \cdot cm^{-2} \cdot d^{-1}$)	mxsol	38	2450	2722.4	3000	131.9
Latitude (°N or °S)	—	38	0.1	45.07	74.4	20.46
Maximum depth (m)	—	34	3.5	32.03	250.0	55.61
Mean depth (m)	Z	38	0.92	12.73	100.00	19.97
Annual phosphorus ($P\text{-}PO_4$, $\mu g \cdot L^{-1}$)	phos	27	1.3	102.06	500.0	137.78
Seasonal chlorophyll ($\mu g \cdot L^{-1}$)	chl	35	0.056	41.11	795.0	137.24
Retention time (yr)	—	18	0.07	1.98	10	3.21
Thermocline depth (m)	—	29	0	6.99	75.3	14.28
pH	—	30	5.8	7.91	10.5	1.01

	Characteristics of zooplankton populations					
	Symbol	N (populations)	Minimum	Mean	Maximum	Standard deviation
Mean annual biomass ($g\ dw \cdot m^{-2}$)	$\bar{B}_{ann}$	108	1.25×10^{-4}	0.311	3.27	0.584
Annual production ($g\ dw \cdot m^{-2} \cdot yr^{-1}$)	P	108	3.86×10^{-3}	11.236	170.13	27.526
Annual P/B	$P/\bar{B}_{ann}$	108	2.63	41.914	220.95	37.425
Mean annual water temperature (°C)	T_{ann}	108	0.9	11.21	26.2	6.94
Mean growing season biomass ($g\ dw \cdot m^{-2}$)	$\bar{B}_{gs}$	87	7.5×10^{-4}	0.505	5.36	0.889
Growing season P/B	$P/\bar{B}_{gs}$	87	1.84	20.216	144.08	22.687
Mean growing season water temperature (°C)	T_{gs}	87	0.9	14.06	27.5	5.87
Growing season duration (days)	gs	87	62	194.7	365	90.8
Mean female adult mass (mg dw)	M_{ad}	86	2.2×10^{-5}	0.0230	0.848	0.0939
Mean individual mass (mg dw)	M_{av}	39	1.04×10^{-5}	7.19×10^{-3}	0.068	0.0141

Note: The symbol column lists the symbol used in the main text to identify variables used in particular analyses.

We feel that such nonlinear effects of population biomass on population production can also be dealt with by including population biomass with the other independent variables in regression analyses of observed variation in weight-specific production rates. This approach yields results that are easy to interpret, plus it has some small claim to greater statistical validity. All regression methods assume that dependent and independent variables are estimated independently of each other (Jackson et al. 1991); however, all methods of estimating production incorporate, in their estimates, the basic data used to estimate biomass. Several commonly used methods (e.g., instantaneous growth method, Rigler and Downing 1984, equation 2.12) are structured as follows: weight-specific production and biomass are estimated independently for some segment of the population (e.g., a particular developmental stage), production for this segment is then calculated by multiplying these two estimates, and total production for the population is estimated as the sum of production estimates for all population segments. Thus, it can be argued that weight-specific production and biomass come closer to the ideal of independently estimated variables than total production and biomass and hence are more appropriate variables to use in regression analyses.

Given this argument and the structure of equation 2, it is apparent that annual production is the product of three potentially independent elements ($[P/B]_{daily}$, $\bar{B}_{gs}$, and gs). Our analyses will focus on these three elements and the various abiotic and biotic factors that may influence them.

Results

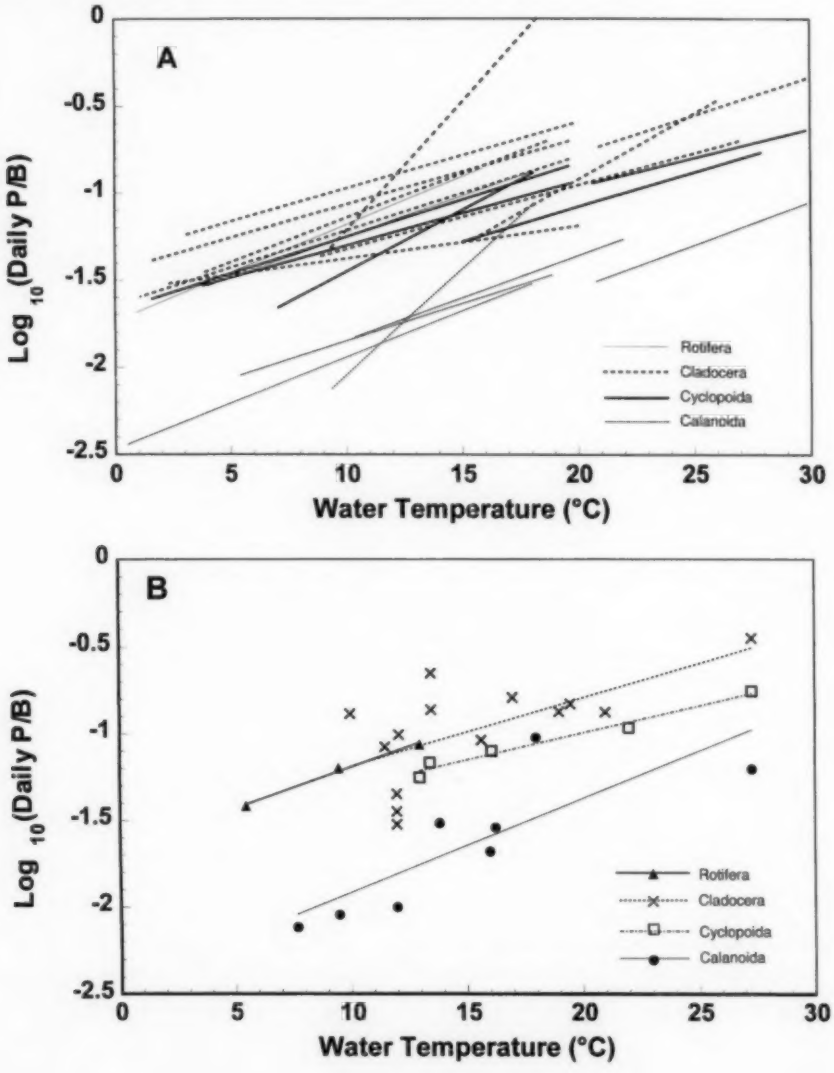
Weight-specific production rates

Daily data set

For 30 populations (25 temperate, 5 tropical) in 18 lakes (16 temperate, 2 tropical), we found detailed information on

© 1997 NRC Canada

Fig. 1. (A) Individual regression lines generated by fitting the model $\log([P/B]_{daily}) = \alpha + \beta(T_{daily})$ to the data for each population found to have a statistically significant ($p < 0.05$) production–temperature relationship. (B) Regression lines generated by fitting eq. 6 ($\log_{10}$(median $[P/B]_{daily}$) = α_{taxa} + β(median T_{daily})) to the median T_{daily} and median $[P/B]_{daily}$ associated with each of the 30 population data sets. The lines are differentiated by taxonomic group.



within-year variation in production, biomass, and water temperature, typically derived from biweekly or monthly sampling programs. Table 3 shows the range of daily water temperatures (T_{daily}) and daily weight-specific production rates ($[P/B]_{daily}$) for the 30 populations in the daily data set. Minimum values for T_{daily} ranged from 0.4 to 20.8°C, and maximum values ranged from 12.8 to 29.8°C. Minimum $[P/B]_{daily}$ values ranged from 0 to 0.169, and maximum values ranged from 0.040 to 0.8400.

Simple regression analysis was used to test each population data set for a positive link between $\log_{10}([P/B]_{daily})$ and T_{daily}. Only data sets with at least six points were included in this analysis. Of the 28 data sets that met this criterion, all exhibited positive correlations and 75% (21) were significant ($p < 0.05$, one-sided test). Of the populations that exhibited significant correlations, about 70% (14) had R^2 values that exceeded 70% and p-values that were less than 0.001 (median p-value = 0.00005). Over 80% had slope estimates ($[P/B]_{daily}$ on temperature) that were similar (i.e., in the range 0.035–0.055) to the estimates (0.040, 0.049) obtained by Huntley and Lopez (1992) for marine copepods (Fig. 1A; Table 4). A metaanalysis of these 28 independent data sets was performed using Fisher's χ^2 test (Sokal and Rohlf 1981; Hedges and Olkin 1985; a procedure that pools the results from several

© 1997 NRC Canada

Table 3. Range (max.–min.) and median of monthly water temperatures and daily *P/B* values for all populations in the daily data set.

T_{daily} range	T_{daily} median	$[P/B]_{daily}$ range	$[P/B]_{daily}$ median	Source
		Significant Cladocera		
9.1	27.3	0.276	0.355	Lévêque and Saint-Jean 1983
17.0	10.0	0.219	0.132	Jones et al. 1979
18.0	21.0	0.210	0.133	Hanazato and Yasuno 1985
8.9	13.5	0.741	0.223	Forsyth and James 1991
14.5	11.5	0.180	0.085	Andrew 1983
10.5	19.5	0.226	0.149	King and Greenwood 1992*b*
18.6	15.6	0.199	0.092	Vijverberg and Richter 1982*a*
18.1	12.1	0.212	0.098	Vijverberg and Richter 1982*a*
		Nonsignificant Cladocera		
17.5	12.0	0.118	0.036	Mason and Abdul-Hussein 1991
17.5	12.0	0.149	0.045	Mason and Abdul-Hussein 1991
9.0	12.0	0.060	0.030	Schindler 1972
1.0	17.0	0.054	0.162	Borsheim et al. 1988
11.0	19.0	0.060	0.134	Babitskiy 1970
8.9	13.5	0.777	0.137	Forsyth and James 1991
		Significant Calanoida		
9.1	27.3	0.063	0.063	Lévêque and Saint-Jean 1983
13.5	16.0	0.045	0.021	Burns 1979
8.9	13.8	0.081	0.034	Forsyth and James 1991
11.5	16.2	0.036	0.029	Green 1976
		Nonsignificant Calanoida		
9.0	12.0	0.011	0.010	Schindler 1972
17.3	11.9	0.064	0.008	Bosselman 1975
7.8	9.5	0.032	0.009	Rey and Capblancq 1975
12.0	18.0	0.060	0.095	King and Greenwood 1992*a*
		Significant Cyclopoida		
9.1	27.3	0.168	0.178	Lévêque and Saint-Jean 1983
13.0	22.0	0.102	0.109	Gophen 1978
11.0	13.0	0.166	0.056	Babitskiy 1988
18.1	13.4	0.215	0.068	Vijverberg and Richter 1982*b*
15.8	16.1	0.123	0.079	Vijverberg and Richter 1982*b*
		Significant Rotifera		
14.0	10.0	0.249	0.059	Andrew and Fitzsimons 1992
15.0	5.5	0.108	0.038	Cajander 1983
		Nonsignificant Rotifera		
13.5	13.0	0.215	0.087	Bosselman 1979*a*, 1979*b*

Note: The populations are grouped by significance ($p < 0.05$) and taxonomic group where significance denotes populations with a statistically significant $[P/B]_{daily}$–T_{daily} relationship. Only time periods with nonzero $[P/B]_{daily}$ values were used in these analyses.

© 1997 NRC Canada

Table 4. Results of fitting the model $\log([P/B]_{daily}) = \alpha + \beta T_{daily}$ to data from individual populations in the Daily Data Set.

Taxonomic group	No. of populations	Individual population analysis			
		N	R^2	α	β
Rotifera	2	22	0.483	−1.748	0.052
		(12, 32)	(0.23, 0.74)	(−1.77, −1.72)	(0.047, 0.056)
Cladocera	8	12	0.818	−1.725	0.044
		(7, 33)	(0.56, 0.99)	(−2.75, −1.40)	(0.037, 0.15)
Cyclopoida	5	12	0.826	−1.766	0.040
		(6, 28)	(0.74, 0.92)	(−2.14, −1.69)	(0.038, 0.069)
Calanoida	4	12	0.780	−2.458	0.050
		(12, 16)	(0.67, 0.94)	(−3.26, −2.26)	(0.042, 0.12)

Note: Results are summarized for those populations that exhibited a statistically significant ($p < 0.05$) temperature effect. Median values and ranges are provided for sample size per population (N), square of the population specific correlation coeficient (R^2), α estimates, and β estimates.

independent tests of an hypothesis to provide a single overall test of that hypothesis) and this indicated that the temperature effect was highly significant ($p < 0.0001$).

Results from the simple regression analyses of $\log_{10}([P/B]_{daily})$ on T_{daily} show that taxonomic groups can be ordered, from high $[P/B]_{daily}$ values to low, as follows: Rotifera/Cladocera, Cyclopoida, Calanoida (Table 4; Fig. 1A).

Although most populations exhibited similar slopes, the two populations from Lake Okaro were extreme exceptions. The temperature coefficients (β) from the two Okaro populations are high compared with others from their respective taxonomic groups. The mean slope for the $[P/B]_{daily}$ versus T_{daily} relationship among the individual cladoceran populations, with the removal of the Okaro populations, is 0.0476, and the Okaro population yields a slope of 0.1488. A similar discrepancy is found within the calanoid group. The mean slope of the individual populations minus the Okaro population is 0.0474, and the Okaro population shows a slope of 0.1203.

For each population, values for $[P/B]_{daily}$ and T_{daily} were derived from observational time series, covering one to three annual cycles. Because the degree of autocorrelation in each time series is relatively high (e.g., r lag $1 \approx 0.7$), the possibility exists that the high and consistent P/B–temperature correlations characteristic of these data sets may reflect concurrent, but unrelated, seasonal variation in the variables rather than a strong, direct association between them. We used the procedure described in Sutcliffe et al. (1976) to remove the influence of autocorrelation from our within-population tests of significance: the number of populations exhibiting significant correlations between $\log_{10}([P/B]_{daily}$ and T_{daily} declined from 75 to 61% and the median p-value declined from 0.00005 to 0.022. However, when we applied Fisher's metaanalysis procedure to these reduced p-values, it still yielded a highly significant result ($p < 0.001$).

Best subsets multiple regression was used to determine if zooplankton biomass exerted a significant influence on the $[P/B]_{daily}$–T_{daily} correlations evident in most populations. Of the 28 populations studied, only four (one cladoceran, one calanoid, and two cycloploid) exhibited statistically significant ($p < 0.05$) biomass effects. In all four populations, biomass had a negative effect on $[P/B]_{daily}$. In three of the populations, the biomass effect was less important (regression mean square for temperature effect 4–20 times regression mean square for biomass effect) than the temperature effect. In only one (Lake Okaro, *Ceriodaphnia dubia*) was biomass more important than temperature in explaining variation in weight-specific production.

The within-population analyses described above exploit the annual variation in temperature characteristic of each lake to estimate the effect of temperature on $[P/B]_{daily}$. There are also large interlake differences in annual temperature range (Table 2), and these differences can be exploited to obtain a second, independent estimate of the effect of temperature on $[P/B]_{daily}$. This was done as follows. Each of the 30 population data sets was characterized by its median T_{daily} value and its median $[P/B]_{daily}$ value. Analysis of covariance (ANCOVA) was then used to fit the following model to these data (Fig. 1B):

$$(6) \quad \log_{10}(\text{median}[P/B]_{daily}) = \alpha_{taxon} + \beta \cdot (\text{median } T_{daily}).$$

The value of β did not vary significantly ($p < 0.05$) across taxonomic groups. Estimates for β (0.04336, SE = 0.00773) and for taxon-specific α (rotifers: −1.631; cladocerans: −1.645; cyclopoids: −1.844; calanoids: −2.294) were all relatively similar to the respective medians of the β and α estimates obtained from the within-population analyses (Table 4).

Annual and growing season data sets

Influence of temperature, population biomass, and body size: We identified useable, annual production estimates for 108 distinct populations found in 38 lakes: 5 arctic lakes with 16 populations, 26 temperate lakes with 73 populations, and 7 tropical lakes with 19 populations. This is our annual data set. Many of these populations were classified to species, but a significant number were only identified to phylum, subclass, or order (i.e., Rotifera, Copepoda, Cladocera). Summary statistics on selected variables are presented in Table 2.

© 1997 NRC Canada

Table 5. Regression results from fitting various combinations of the model $\log(P/B_{ann}) = \alpha_{taxon} + \beta(T_{ann}) + \delta(\log\overline{B}_{ann}) + \tau(\log M_{ad})$ where α_{taxon} is an additive constant specific to each taxonomic group.

Regression statistics	Independent variables included in regression			
	T_{ann}, B_{ann}	T_{ann}, M_{ad}	T_{ann}, B_{ann}, taxa	T_{ann}, B_{ann}, taxa refined
N	108	86	108	90
R^2_{adj}	0.270	0.212	0.522	0.572
$\alpha_0 \pm$ SE	0.960 ± 0.088	0.954 ± 0.115	—	—
$\alpha_{rot} \pm$ SE	—	—	1.195 ± 0.097	1.355 ± 0.057
$\alpha_{cal/chy} \pm$ SE	—	—	1.212 ± 0.079	1.288 ± 0.066
$\alpha_{cyc} \pm$ SE	—	—	0.888 ± 0.089	0.963 ± 0.073
$\alpha_{cal} \pm$ SE	—	—	0.710 ± 0.083	0.680 ± 0.083
$\beta \pm$ SE	0.030 ± 0.005	0.025 ± 0.006	0.028 ± 0.004	0.029 ± 0.004
$\delta \pm$ SE	–0.154 ± 0.035	—	–0.080 ± 0.033*	—
$\tau \pm$ SE	—	–0.094 ± 0.031	—	—

Note: α_0 = estimate obtained when data from all taxonomic groups are pooled; α_{rot}, Rotifera; $\alpha_{cal/chy}$, Cladocera/Chydoroidea; α_{cyc}, Cyclopoida; α_{cal}, Calanoida. *$0.05 > p > 0.01$.

Of the 108 populations in the annual data set, there were 105 with the additional data necessary for estimating length of growing season. Of these 105, 87 could be classified into taxonomically similar groups with relatively large (>10) sample sizes per group. These 87 populations represent the same 38 lakes as were included in the annual data set; however, the numbers of populations in arctic, temperate, and tropical lakes are reduced to 15, 61, and 11, respectively. Four taxonomic groups are represented: (*i*) Chydoroidea, 30 data sets; (*ii*) Calanoida, 14 data sets; (*iii*) Cyclopoida, 20 data sets; and (*iv*) Rotifera, 23 data sets. Populations classified at higher taxonomic levels (i.e., copepods, cladocerans) were omitted in order to insure some minimal level of similarity among the organisms included within each group. The number of groups was kept at four to insure a reasonable sample size within each group. For example, we chose to limit our analysis of cladocerans to the superfamily Chydoroidea (or Daphnoidea, Hutchinson 1967) consisting of the five families Chydoridae, Macrothricidae, Bosmidae, Moinidae, and Daphniidae because it provided a good sample size (30) of species that were relatively homogeneous in terms of their morphological characteristics. Sample sizes in other cladoceran groups were small (Haplopoda, 1; Sidoidea, 6; Polyphemoidea, 1) and the species represented typically exhibited very distinctive characteristics (e.g., the very large body size of *Leptodora kindtii*; the gelatinous sheath of *Holopedium* spp.). The 87 populations, selected as described above, make up our growing season data set (Table 1).

We begin by examining whether the proportion of observed variation in $P/\overline{B}_{ann}$, associated with variation in adult body size (Plante and Downing 1989), can also be accounted for by splitting the $P/\overline{B}_{ann}$ values into subsets based on taxonomy. First, models for $P/\overline{B}_{ann}$ were generated (Table 5) based on T_{ann} and either M_{ad} or $\overline{B}_{ann}$ because the R^2 for a (T_{ann}, M_{ad}, $\overline{B}_{ann}$) model was not significantly greater than the R^2 for a (T_{ann}, M_{ad}) model or a (T_{ann}, $\overline{B}_{ann}$) model. The fits of these models were inferior to the fits of models that took taxonomic groupings into account. With taxonomy included in the regression model, the influence of body size disappeared and the influence of population biomass was weakened (Table 5). When the analysis was further refined to include only the relatively homogeneous taxonomic groups identified for the growing season data set, the biomass effect disappeared and the overall fit of the model improved (Table 5). Removing nonsignificant terms, we obtain the following final model ($N = 90$, $R^2_{adj} = 0.5731$) for our annual data set (Fig. 2):

$$(7) \quad \log_{10}(P/\overline{B}_{ann}) = \alpha_{taxon} + 0.0293 \cdot T_{ann}$$

where α_{taxon} equals 1.33 (SE = 0.051) for Rotifera and Chydoroidea, 0.975 (SE = 0.072) for Cyclopoida, and 0.692 (SE = 0.082) for Calanoida.

With these results in mind, we then examined the ability of our growing season model (equation 5) to describe the growing season data set. The following equation was fitted to the data:

$$(8) \quad \log_{10}(P/\overline{B}_{gs}) = \alpha + \beta \cdot T_{gs} + \Phi \cdot \log_{10}(gs)$$

where α could be influenced by taxonomy, $\overline{B}_{gs}$, M_{ad}, and M_{av}. We used ANCOVA and best subsets regression analysis on the growing season data set to test the usefulness of this model in accounting for the observed variation in $P/\overline{B}_{gs}$. We found that $P/\overline{B}_{gs}$ varied significantly with taxonomic group, T_{gs}, and gs (Table 6). As expected, both of these effects were positive. The temperature effect was similar across taxonomic groups (ANCOVA tests for interaction effects between taxonomic group and temperature were not significant: $p > 0.05$), and at a given temperature, $P/\overline{B}_{gs}$ declined with taxonomic group in the following sequence: Rotifera, Chydoroidea, Cyclopoida, Calanoida (Table 6); $\overline{B}_{gs}$, M_{ad}, and M_{av} did not significantly influence $P/\overline{B}_{gs}$. Identical results were obtained with a similar analysis using $\log_{10}P$ as the dependent variable: the regression coefficient for $\log_{10}\overline{B}_{gs}$ was not significantly different from 1,

© 1997 NRC Canada

Fig. 2. Plot of annual P/B and mean annual water temperature estimates. The lines represent the regression model $\log(P/\bar{B}_{ann}) = \alpha_{taxon} + 0.029(T_{ann})$ for the refined taxonomy data set ($N = 90$), where α_{taxon} is an additive constant specific to each taxonomic group: α_{rot}, Rotifera = 1.355; α_{chy}, Chydoroidea = 1.288; α_{cyc}, Cyclopoida = 0.963; α_{cal}, Calanoida = 0.680.

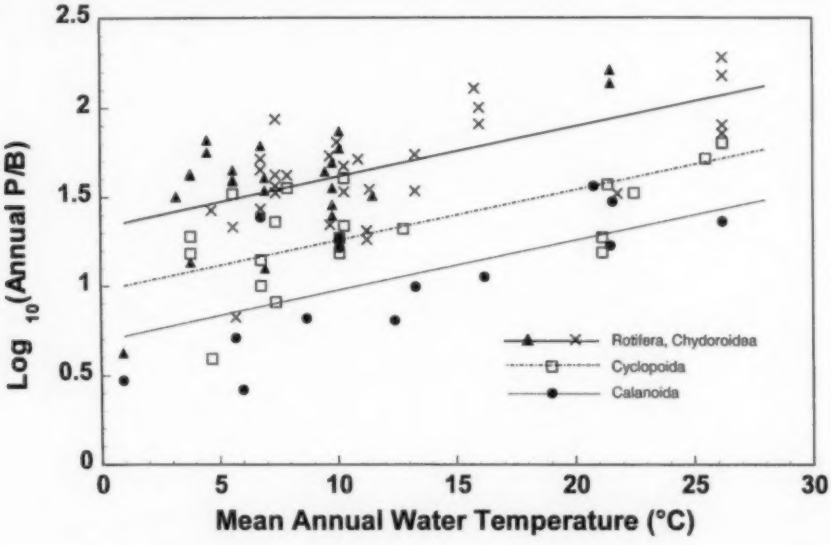


Table 6. Comparison of regression results obtained by fitting the model $\log(P/\bar{B}_{gs}) = \alpha_{taxon} + \beta(T_{gs}) + \Phi(\log(gs))$ to our most comprehensive growing season data set and to two refined subsets of the data where α_{taxon} is an additive constant specific to each taxonomic group.

Regression statistics	Complete growing season dataset	Subset 1	Subset 2
N	87	64	45
R^2_{adj}	0.700	0.764	0.768
$\alpha_{rot} \pm$ SE	−0.634 ± 0.267	−0.637 ± 0.266	−0.809 ± 0.440
$\alpha_{chy} \pm$ SE	−0.784 ± 0.262	−0.803 ± 0.267	−0.956 ± 0.404
$\alpha_{cyc} \pm$ SE	−1.019 ± 0.281	−0.892 ± 0.291	−1.197 ± 0.451
$\alpha_{cal} \pm$ SE	−1.191 ± 0.295	−1.200 ± 0.303	−1.479 ± 0.486
$\beta \pm$ SE	0.042 ± 0.004	0.045 ± 0.004	0.042 ± 0.006
$\Phi \pm$ SE	0.629 ± 0.124	0.600 ± 0.124	0.712 ± 0.209

Note: α_{rot}, Rotifera; α_{chy}, Chydoroidea; α_{cyc}, Cyclopoida; α_{cal}, Calanoida. Subset 1 represents populations in which water temperatures were directly measured; subset 2 represents those production estimates that were derived using methods that did not assume food-saturated development times.

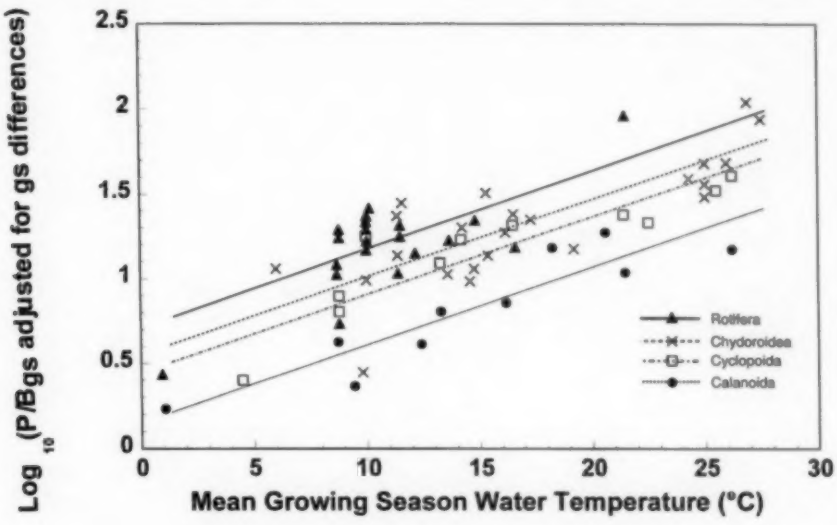
the coefficient for T_{gs} was similar for all taxonomic groups, and the group effects themselves were similar to those identified in the $P/\bar{B}_{gs}$ analysis.

Given the overriding importance of temperature in the growing season data set, we examined the possibility that a more accurate model could be developed from that portion of the data set where T_{gs} was estimated directly from water temperature data provided in the original references. Analysis of this subset of 64 cases does produce a model that fits the data better: the structure of the best model was identical with that fitted to the full growing season data set, but its residual mean square was smaller (0.033 versus 0.040) and its R^2 value was higher (Table 6; Fig. 3). In addition, this increase in accuracy is not accompanied by a reduction in the range of values for the independent variables over which the model can be legitimately applied: the ranges for the independent variables in the reduced data set were identical with the ranges found in the full growing season data set.

The structure of eq. 5 implies that Φ (eq. 8) should have a value close to 1. Our best estimate for Φ (0.6054, SE = 0.122) is close to, but significantly less than, 1. A plot of standardized residuals against log(gs) exhibited strong negative curvature at high (>2.45) $\log_{10}$(gs) values. If Φ is reestimated (ANCOVA of cases with direct T_{gs} estimates) from that subset

© 1997 NRC Canada

Fig. 3. Plot of corrected $P/\overline{B}_{gs}$ values against mean growing season water temperature estimates: corrected $P/\overline{B}_{gs}$ = observed $P/\overline{B}_{gs} - 0.600(\log(gs) - \text{mean } \log(gs))$, where mean log(gs) = 2.2423 and 0.600 comes from the regression model $\log(P/\overline{B}_{gs}) = \alpha_{taxon} + 0.045(T_{gs}) + 0.600(\log(gs))$ fitted to the refined taxonomy data set ($N = 64$). The lines represent the same model where α_{taxon} is an additive constant specific to each taxonomic group: α_{rot}, Rotifera = −0.637; α_{chy}, Chydoroidea = −0.803; α_{cyc}, Cyclopoida = −0.892; α_{cal}, Calanoida = −1.200.



($N = 49$) of the data with gs values less than 300, the new estimate (0.800, SE = 0.17) is not significantly different from 1. Similar results are obtained when cases with both direct and indirect T_{gs} estimates are included in the analysis. These results strongly suggest that our procedure for estimating gs is biased high for estimated values greater than 300 days. In fact, the distribution of gs estimates is strongly bimodal: 83% have values less than 300 days and are normally distributed around a mean value of 160 days; the remaining 17% form a relatively uniform subset, with almost all values (12 of 15) equal to 365 days.

A rough estimate of the bias associated with this subset of the data can by obtained by refitting the regression models of Table 6, with Φ set to 1.0 and a new categorical variable (long gs) added that splits the data into two subsets at a gs value of 300 days. The regression coefficient estimated for long_gs then provides a direct measure of the potential bias associated with gs estimates greater than 300 days. These new models provide improved descriptions of their respective data sets (i.e., small increases in R^2_{adj} values). In both, long_gs is statistically significant ($p < 0.0001$) and has fitted values indicating that gs estimates in the neighbourhood of 365 days overestimate the "true" gs values by about 60%. Also, by fixing Φ at 1, estimates of α_{taxon} and β derived from the growing season data sets can be compared directly with estimates of α_{taxon} and β obtained by fitting eq. 6 to the $[P/B]_{daily}$ data (Table 7). The similarity among all three sets of estimates confirms the prediction, inherent in eq. 4, that the relationship linking $[P/B]_{daily}$ values to daily temperatures can be recovered by fitting eq. 5 to $P/\overline{B}_{gs}$ values.

Influence of food resource availability and lake morphometry: All subsets regression analysis and stepwise regression analysis were used to determine if addition of the independent variables phos, chl, and *Z* to the models listed in Table 6 would produce a significant ($p < 0.05$) increase in their adjusted R^2 values. No such increase was observed. Therefore, we conclude that none of these variables improves on the explanatory power of the models in Table 6.

Comparing empirical predictors of production: We compared the predictive capability of our "best" taxonomically differentiated model for annual production:

$$(9)\quad \log_{10}(P) = \alpha_{taxon} + \log_{10}(\overline{B_{gs}}) + 0.045 \cdot T_{gs} + 0.600 \cdot \log_{10}(gs)$$

(where $\alpha_{rot} = -0.634$, $\alpha_{chy} = -0.784$, $\alpha_{cyc} = -1.019$, and $\alpha_{cal} = -1.191$) with the Plante and Downing (1989) annual production model:

$$(10)\quad \log_{10}(P) = 0.06 + 0.79 \cdot \log_{10}(\overline{B_{ann}}) - 0.16 \cdot \log_{10}(W_m) + 0.05 \cdot T_{ann}$$

where $\overline{B}_{ann}$ = mean annual biomass (grams dry weight per square metre), W_m = maximum adult size, and T_{ann} = mean annual water temperature (degrees Celsius). The comparison was limited to the subset of 48 cases that contained all variables necessary for applying both equations. For our growing season model, the R^2 calculated from observed and predicted values for $\log_{10}(P)$ was 0.9521, the mean residual was −0.0093, and the standard deviation of the residuals was 0.2087. Predictions derived from the Plante and Downing (1989) model were not as accurate: the R^2 was 0.8679, the mean residual was 0.1928, and the standard deviation of the

© 1997 NRC Canada

Table 7. Comparison of three sets of estimates for the relationship $\log([P/B]_{daily}) = \alpha_{taxon} + \beta \cdot \log(T_{daily})$ where α_{taxon} is an additive constant specific to each taxonomic group.

Parameters	Estimates from daily data set	Estimates from growing season data set	
		Subset 1	Subset 2
N	30	64	87
R^2_{adj}	0.737	0.776	0.704
$\alpha_{rot} \pm$ SE	−1.631 ± 0.141	−1.482 ± 0.058	−1.424 ± 0.060
$\alpha_{cla} \pm$ SE	−1.645 ± 0.132	−1.651 ± 0.084	−1.565 ± 0.085
$\alpha_{cyc} \pm$ SE	−1.844 ± 0.170	−1.732 ± 0.093	−1.825 ± 0.088
$\alpha_{cal} \pm$ SE	−2.294 ± 0.138	−2.047 ± 0.096	−2.005 ± 0.094
$\beta \pm$ SE	0.0434 ± 0.008	0.0451 ± 0.004	0.0419 ± 0.004

Note: α_{rot}, Rotifera; α_{cla}, Cladocera; α_{cyc}, Cyclopoida; α_{cal}, Calanoida. The estimates in the first column were obtained by fitting eq. 6 to the daily data set. Estimates derived from the growing season data set were obtained by fitting eq. 8, with Φ set to 1 and the long gs term included. Subset 1 was derived from T_{gs} estimates based on direct water temperature measurements; subset 2 was derived using all available estimates of water temperatures. Coefficient estimates for the long gs term were −0.22 (SE = 0.062) for subset 1 and −0.172 (SE = 0.063) for subset 2.

residuals was 0.3237. When we confined our comparison to those cases ($N = 28$) with direct estimates of temperature, predictions from the growing season model ($R^2 = 0.9803$, mean residual = −0.0468, standard deviation of residuals = 0.1740) were still more accurate than those derived using the Plante and Downing (1989) model ($R^2 = 0.8865$, mean residual = 0.1037, standard deviation of residuals = 0.3253).

Average biomass over the growing season

Stepwise regression analysis produced the following empirical model for $\overline{B}_{gs}$:

$$(11)\quad \log_{10}(\overline{B}_{gs}) = -0.139 + 0.538 \cdot \log_{10}(M_{ad}) + 0.346 \cdot \log_{10}(\text{chl}) + 0.412 \cdot \log_{10}(Z)$$

($N = 71$, $R^2\text{adj} = 0.580$) with all terms highly significant ($p < 0.01$). In this analysis, about 48% of the overall variation in $\log_{10}(\overline{B}_{gs})$ was accounted for by $\log_{10}M_{ad}$, 6% was accounted for by $\log_{10}$chl, and 4% by $\log_{10}Z$. The group of observations with long (>300 days) gs values did not stand out from all the other data.

Length of the growing season

Stepwise regression analysis produced the following empirical model for gs:

$$(12)\quad \log_{10}(\text{gs}) = 2.16 + 0.165 \cdot \log_{10}(\text{chl}) - 0.059 \cdot (\log_{10}(\text{chl}))^2 - 0.113 \cdot \text{clad} + 0.334 \cdot \text{long gs}$$

($N = 85$, $R^2\text{adj} = 0.61$) with all terms highly significant ($p < 0.001$). In this analysis, about 10% of the overall variation in $\log_{10}$(gs) was accounted for by the chl terms, 14% was accounted for by the clad term, and 37% by the long gs term. The value (0.344, SE = 0.038) of the long gs term in this model is not significantly ($p > 0.05$) different from the estimate (0.221, SE = 0.062) of its value obtained from the $P/\overline{B}_{gs}$ model (Table 7). Thus, both analyses (i.e., $P/\overline{B}_{gs}$ and gs) generate similar estimates for the positive bias present in gs estimates greater than 300 days.

Discussion

Is the association between weight-specific production and water temperature a methodological "artifact"?

All our analyses demonstrate a strong and consistent correlation between water temperature and weight-specific production. This could be the product of the direct effects of temperature on the biological processes of production or it could simply reflect the fact that "adjustments" for temperature were included in the calculations used to generate the original production estimates. One of the most common methods for estimating production assumes that individual development rates in the field match food-saturated development rates measured in the laboratory (Rigler and Downing 1984). This assumption is included in production calculations by estimating field development rates from field water temperatures using temperature functions defined in the laboratory under food-saturating conditions. Although this assumption may be generally true for the development times of life stages that do not feed independently (eggs, Edmondson 1974 and references therein; early naupliar stages, Hart 1990 and references therein), there is good evidence that the development times of life stages that do feed independently (later naupliar and copepodite stages) are significantly extended by large reductions in food supply (Rigler and Downing 1984; Hart 1990; McCauley et al. 1990*a*). If a large percentage of authors generated their production values from individual development times estimated from field water temperatures, then these production values should exhibit strong dependence on field water temperatures, everything else being equal. The presence of such an effect would be a simple consequence of the

© 1997 NRC Canada

methodological assumption of the authors, and would in no way provide independent evidence of the validity of that assumption.

To assess this possibility, we identified those production estimates in our data set that were free of this assumption and then examined the character of the temperature effect exhibited by those estimates alone. The methods in our data set fall into two broad categories: (*i*) those based on growth rate estimates for individual organisms (increment–summation and related methods, Waters 1977) and (*ii*) those based on population growth rate estimates, including turnover time methods and removal–summation methods (Waters 1977).

Among studies using type *i* methods, in 15, development times were estimated directly in the field by timing the passage of individual cohorts through their developmental stages; in 13, field development times were assumed equal to laboratory-estimated development times measured under food, water quality, and temperature conditions set to match those existing in the field; and in 16, field development times were assumed equal to laboratory-estimated development times measured under food-saturating conditions at field temperatures.

Among studies using type *ii* methods, 17 used nonequilibrium variants of the removal–summation approach, where production estimates are generated from direct estimates of short-term variation in population growth rate in the field, coupled with indirect estimates of population birth rate from egg densities and estimated egg development times. Typically, egg development times were estimated from field water temperatures using a laboratory-derived function. Such methods generate production estimates that should be quite comparable with those generated by type *i* methods (Crisp 1984; Gillespie and Benke 1979; Herzig et al. 1980).

Consider our analyses of the $[P/B]_{daily}$ data. Of the 28 data sets analyzed, six were based on direct estimates of field development times. In all six, $[P/B]_{daily}$ was positively correlated with T_{daily}. In three, the correlation was statistically significant at the 1% level, and in the remaining three, it was significant at the 10% level. In regression analyses of these data sets, the median value for the temperature coefficient (0.055) was similar to the value derived from the entire daily data set (0.0434 ± 0.008, Table 7). A further six data sets were based on development times estimated from laboratory simulations of field conditions: all six exhibited positive and statistically significant ($p < 0.01$) correlations between $[P/B]_{daily}$ and T_{daily}, with the median estimate (0.040) of the temperature coefficient again very similar to the value derived from the entire data set. Thus, the finding of a strong and consistent association between daily, weight-specific production rates and temperature does not simply reflect assumptions buried within the production estimation methods.

Now consider our analyses of $P/\overline{B}_{gs}$ values. Of the 87 estimates in the original data set, 15 were based on direct estimates of field development times. This subset of the complete data set exhibited a statistically significant ($p < 0.02$) association with water temperature; the estimate for the temperature regression coefficient (0.0421 ± 0.017) was essentially identical with the estimate derived from the entire data set (0.0425 ± 0.004). An additional 13 estimates were based on development rates derived from laboratory simulations (food availability, water quality, temperature) of field conditions. In this larger subset ($N = 28$) of the data, the temperature effect was again statistically significant ($p < 0.0001$) and the regression coefficient estimate was very similar (0.0451 ± 0.007) to the estimate derived from the entire data set. In addition, this subset exhibited ($p < 0.05$) the difference, between calanoid species and other species, that was strongly apparent in the entire data set. Finally, consider the larger subset ($N = 45$) of data, obtained by including the 17 production estimates derived from removal–summation methods. Analysis of these data produced a regression model that was essentially identical with the model generated by our analysis of the entire data set (Table 6). We used this model (Table 6, subset 2) to predict $P/\overline{B}_{gs}$ values for the 16 studies that were explicitly based on the assumption that field development times equalled temperature-corrected food-saturated development times. There was a strong association ($r = 0.8$, $p < 0.001$) between predicted and observed values and no sign of systematic bias in the predicted values: the regression of observed values on predicted values had slope and intercept estimates not significantly ($p > 0.05$) different from 1 and 0, respectively. These findings show that the character of the temperature effect resident in the growing season data set is not a product of methodological assumptions buried in the production calculations themselves.

Our analyses of daily and annual production data suggest that simple strong relationships, linking weight-specific production rate to temperature, underlie a substantial portion of the variation in production observed in natural populations of limnetic zooplankton. When we examined variation in $[P/B]_{daily}$ values within populations, we found that 75% of the populations examined exhibited such relationships. The similarity of slope estimates and the distinctiveness of intercept estimates (Table 4; Fig. 1) clearly characterize the ways in which different taxonomic groups vary in terms of both their characteristic daily weight-specific production values and the sensitivity of those values to changes in temperature. A second independent analysis, founded on between-population differences in $[P/B]_{daily}$ and T_{daily} values, produced a quantitatively similar picture (Fig. 1). A third independent analysis, founded on between-population differences in annual production values, produced a picture that was both qualitatively and quantitatively consistent (Figs. 2 and 3; Table 7) with the results obtained from the analyses of $[P/B]_{daily}$ values.

All of these results are consistent with the view that the combined influence of temperature and taxonomy on annual production rates in the wild is strong, pervasive, and accurately described by a model of the following form:

$$(13)\quad \log_{10}(P) = \alpha_{taxon} + \log_{10}(\overline{B}_{gs}) + \beta\cdot\overline{T}_{gs} + \Phi\cdot\log_{10}(gs)$$

or

$$(14)\quad \log_{10}(P/\overline{B}_{gs}) = \alpha_{taxon} + \beta\cdot T_{gs} + \Phi\cdot\log_{10}(gs)$$

where α varies with taxonomic group.

Our most accurate version of this model (Table 6, direct T_{gs} estimates) has the following properties: (*i*) it accounts for a very large (77%) proportion of observed variation in $P/\overline{B}_{gs}$ values; (*ii*) α varies significantly among taxonomic groups, reflecting the same ordering of production rates (highest to lowest: Rotifera, Cladocera/Chydoroidea, Cyclopoida, Calanoida) observed in our analysis of the daily production data; and (*iii*) variation in β (0.0458, SE = 0.0039) across taxo-

© 1997 NRC Canada

nomic groups was undetectable; the estimated value for β was very similar to independent estimates derived from analyses of $[P/B]_{daily}$ values (i.e., pooled β value from between-population analysis = 0.04336, SE = 0.0077; median β values from within-population analyses: Rotifera, 0.0516; Cladocera, 0.0442; Cyclopoida, 0.0399; Calanoida, 0.0499).

Our estimate for β is very similar to recent β estimates (0.048 ± 0.001; 0.041 ± 0.005) reported by Huntley and Lopez (1992) for marine copepods. It also reflects a temperature effect that is quantitatively similar to the temperature effect evident in Hart's (1990) compilation of individual development times among marine and freshwater copepods, under food-saturated conditions. Regression analyses of these data, assuming a Van't Hoff temperature relationship (i.e., eq. 1), generated temperature coefficient estimates (freshwater calanoids, 0.039 ± 0.002; marine calanoids, 0.041 ± 0.004; freshwater cyclopoids, 0.048 ± 0.006) that overlap our β estimates and those of Huntley and Lopez (1992). In addition, some of the differences in development time between subsets of Hart's (1990) data are qualitatively similar to differences in weight-specific production rates noted in our analyses: (*i*) the weight-specific production rates for freshwater calanoids in our data set were substantially lower than those noted for marine copepods by Huntley and Lopez (1992); similarly, in Hart's (1990) data, development times for freshwater calanoids are significantly ($p < 0.01$) longer (by about 65%) than those for marine calanoids; and (*ii*) in our data set, calanoid weight-specific production rates are significantly ($p < 0.05$) lower than cyclopoid rates; similarly, in Hart's (1990) data, freshwater calanoid development times are significantly longer (by about 17%) than freshwater cyclopoid development times.

Associations among population density, food resource availability, body size, and lake mean depth

Many authors have reported a positive association between chlorophyll concentration and total biomass density of zooplankton. Our analysis of population-level biomass density (grams dry weight per square metre of lake surface area) also revealed a positive link to chlorophyll concentration, with our slope estimate for chl (0.35, SE = 0.090, in our log–log regression of $\overline{B}_{gs}$ on chl) not significantly different ($p > 0.05$) from estimates obtained in earlier studies of total zooplankton biomass density: 0.51 (McCauley and Kalff 1981), 0.43–0.53 (Hanson and Peters 1984), and 0.47 (Cyr and Pace 1993).

Empirical studies of a wide range of organisms (Damuth 1981; Peters 1983; Strayer 1994) have reported statistical associations between population density and individual body size. Typically, numerical density declines with increasing body size, and biomass density increases. Both types of relationships are allometric with characteristic body weight exponent values of –0.75 for numerical density and +0.25 for biomass density (Blackburn et al. 1993). The few reported studies of zooplankton (or related organisms) are consistent with this general finding. Peters and Wassenberg (1983) looked at the biomass density of invertebrate populations and found a positive allometric relationship, with a body weight exponent of +0.45. Cyr and Pace (1993) looked at the total biomass density of zooplankton communities and found a positive allometric relationship with mean community body weight (exponent value = +0.75), coupled with a positive relationship with chlorophyll concentration. Our analysis of population biomass density also revealed an allometric relationship with body weight. Our estimate for the weight exponent (0.53, SE = 0.070) was somewhat lower than that found by Cyr and Pace (1993) but somewhat higher than its characteristic value of 0.25. This finding is consistent with the fact that numerical density is allometrically related to body size in our data set, with an exponent value (−0.25, SE = 0.12) that is also higher than its characteristic value (−0.75).

The presence of body size and chlorophyll terms in our biomass regression equation (eq. 11) is consistent with the hypothesis (Damuth 1981; Lawton 1989; Blackburn et al. 1993) that population density – body size correlations largely reflect the reciprocal effects of food availability and individual food requirements on population carrying capacity: more food can support more animals of any size, and because larger animals often have lower weight-specific energy requirements than smaller animals and larger animals may acquire a greater proportion of available resources than smaller animals, a fixed amount of food will usually support a greater biomass density of larger animals.

The positive link between population biomass density (measured as grams dry weight per square metre of lake surface area) and lake mean depth is likely a simple result of the increases in average water volume per unit surface area that accompany increases in lake mean depth.

Much recent work (e.g., McQueen et al. 1989; Carpenter and Kitchell 1993) has shown that the activity of both predators and competitors can strongly influence zooplankton biomass density in lakes, at both the community level and the population level. Because we were unable to capture these potentially important effects in our analyses, it is not surprising that we could only account for a moderate amount (58%) of the observed variation in population biomass density. Also, in our analysis, variation in adult body size accounted for a larger share of observed biomass variation than did variation in chlorophyll concentration. This is partly a reflection of the fact that adult body size exhibits greater variation in our data set than does chlorophyll concentration.

Interpreting the associations between weight-specific production, population biomass, and body size

Our analysis of annual production rates suggests that the statistical associations linking weight-specific production rates to both population biomass and adult body size may simply reflect large, roughly parallel taxonomic differences in all three variables rather than direct influences of population biomass and adult body size on production rates. This conclusion is supported by the following observation. Ordering (from lowest to highest) our four taxonomic groups by median population biomass yields the same sequence (Rotifera, Cyclopoida, Chydoroidea, Calanoida) as ordering them by median adult body weight. A similar sequence (Rotifera, Chydoroidea, Cyclopoida, Calanoida) is obtained if the groups are ordered from the highest weight-specific production rate to the lowest. The similarity in these three separate orderings should produce relatively strong negative correlations between production rate and adult body weight and between production rate and population biomass, if all the data are pooled and analyzed without recognizing taxonomic groupings. This is exactly what we observed in our analyses of the pooled pro-

© 1997 NRC Canada

duction data and what Plante and Downing (1989) observed in their analyses.

There are also process-level interpretations (Peters 1983; Downing 1984) of such empirical associations. Negative effects of population biomass could be interpreted in terms of density-dependent growth processes operating at the population level, and negative effects of body size could be interpreted in terms of the size dependence of energy metabolism (e.g., respiration, maximum growth rate: Peters 1983; Schmidt-Nielsen 1984) commonly observed at the individual level. Our analysis of production data from limnetic zooplankton populations is not consistent with either of these interpretations: (*i*) if our entire zooplankton $P/\overline{B}_{ann}$ data set is analyzed without accounting for the distribution of observations among taxonomic groups, we obtain a strong biomass effect; (*ii*) if we conduct a similar analysis on a reduced data set containing only data from the smallest (Rotifera) and largest (Copepoda) individual sized groups, we obtain a strong body size effect and no biomass effect ($\log_{10}(P/\overline{B}_{ann})$ on T_{ann}, $\log_{10}(M_{ad})$; $R^2 = 0.525$); (*iii*) if we account for taxonomic groupings explicitly in our statistical model, then both the biomass and body size effects disappear, overall model fit improves (Table 5), and similar results are obtained no matter what the distribution of observations among taxonomic groups; and (*iv*) analyses of within-population variation in $[P/B]_{daily}$ values showed that negative effects of population biomass were rarely detectable.

We reanalyzed the original Plante and Downing (1989) set of production data for freshwater invertebrates and also found that results differed depending on how internal groupings within the data were recognized: (*i*) with all data pooled, regression analysis identified strong negative biomass and body size effects on total production; (*ii*) if the analysis was limited to the benthic data alone, the body size effect disappeared and the biomass effect weakened; and (*iii*) if the analysis was limited to the zooplankton data alone, both biomass and body size effects were weakened; however, the continuing presence of the body size effect was due to the presence of a single representative of the Mysidacea in the data set, an organism with a body mass at least 100 times larger than almost all other organisms present; with this observation eliminated, the body size effect disappeared.

Given a functional interpretation of the biomass and body size effects, one would expect that a statistical analysis that allowed for taxonomic differences would permit such effects to emerge more clearly. One would also expect to see strong evidence for density-dependent effects on production in analyses of the seasonal cycle of daily production rates. Neither of these expectations was met in our work.

The absence of body size effects on population *P/B* values is consistent with the following observations on zooplankton: (*i*) among cladocerans (Paloheimo et al. 1982; McCauley et al. 1990*a*, 1990*b*; Glazier and Calow 1992; Paloheimo et al. 1982) and copepods (van den Bosch and Gabriel 1994), weight-specific rates of metabolism, ingestion, and assimilation are almost independent of body mass; (*ii*) among cladocerans, intrinsic rates of population increase (Lynch 1980) and population *P/B* (Banse 1982) values, observed under laboratory conditions, are almost independent of adult body size; (*iii*) regression analysis of data in Hart (1990) shows that life cycle development times (food saturating conditions) for marine and freshwater copepods are strongly dependent on temperature and independent of adult body size; (*iv*) Huntley and Lopez (1992) found that weight-specific rates of production by marine copepods were strongly dependent on temperature but were essentially independent of adult body size; and (*v*) in copepods, individual growth over the life cycle can often be approximated by a simple exponential model (Huntley and Lopez 1992); given this, age structure effects on population biomass growth should be minimal.

Because many of the processes that contribute to population production do not exhibit the size dependence characteristic of other animal groups, it is perhaps to be expected that population *P/B* values for cladocerans and copepods do not exhibit the size dependence characteristic of other animal groups.

Biomass stability and the length of the growing season

Our protocols for estimating gs appear to yield positively biased estimates as gs approaches its upper bound of 365 days. For all populations with gs values >300 days (the long gs group), gs was estimated from the annual biomass cycle. Given our protocol for estimating gs, a relatively stable (i.e., minimum annual biomass >20% maximum annual biomass) biomass cycle guarantees a gs estimate of 365 days. The positive bias in such estimates suggests that a stable annual biomass cycle does not provide useable information on the duration of the productive period for the population. This would arise if biomass stability resulted from mortality rates low enough to prevent rapid loss of accumulated biomass. Cyclopoids, and particularly calanoids, are overrepresented in the long gs group.

Population production and food resource availability

Our analyses suggest that the average durations of developmental stages in the field are not very different from food-saturated values observed in the laboratory. However, this finding does not remove the possibility that field production rates are influenced by food availability. Stage duration is a relatively insensitive indicator of food-limited growth: quite large reductions in food availability are required to produce even moderate increases in duration (Ban 1994; Jamieson and Burns 1988; Makarewicz and Likens 1979). Thus, production rates could be suffering from moderate levels of food limitation while still exhibiting stage durations similar to those characteristic of food-saturated conditions. Production estimates would reflect the effects of food limitation through more sensitive population parameters such as body size (Geller 1989; Hart 1991; Lynch 1989; McCauley et al. 1990*a*, 1990*b*; Santer and van den Bosch 1994) and particularly egg production (Ban 1994; Lynch 1989; Makarewicz and Likens 1979), both of which are usually measured directly in the field.

Our examination of within-population variation in $[P/B]_{daily}$ values suggests that compensatory links between population biomass and production are rarely strong enough to be detectable. Few populations (four, less than 20% of those examined) exhibited such relationships, and in only one was this negative effect of population biomass on production strong (i.e., of more importance than temperature in explaining variation in production) and consistently expressed throughout the growing season.

© 1997 NRC Canada

Our examination of growing season production rates indicates that the growing season average for $[P/B]_{daily}$ is independent of $\overline{B}_{gs}$. This suggests that if strong intraspecific effects of population density on daily weight-specific production rates do occur, they must be restricted to relatively short segments of the growing season.

We were unable to use our data set to test for the effects of interspecific competition on population-specific production rates because most studies did not report biomass levels for other segments of the zooplankton community.

Taken together, our empirical analyses of the elements of the annual production cycle for limnetic zooplankton suggest that (*i*) weight-specific rates of production during the cycle are primarily driven by temperature; the influence of food resource availability is probably only evident over relatively short portions of the cycle; and (*ii*) the level of biomass reached during the cycle is influenced by food resource availability and adult body size; temperature exerts little influence on these elements of the cycle.

This view of the production cycle is similar to that reached by Geller (1987), after his study of zooplankton populations in Lake Constance: "... juvenile development was prolonged by up to a factor of 2 with low food concentrations. In natural populations, however this may be of minor importance, because nearly no offspring is produced during times of severe food shortage (clear-water phase; winter) and because the bulk of a population's annual production occurs under optimum food conditions during the spring maximum of phytoplankton."

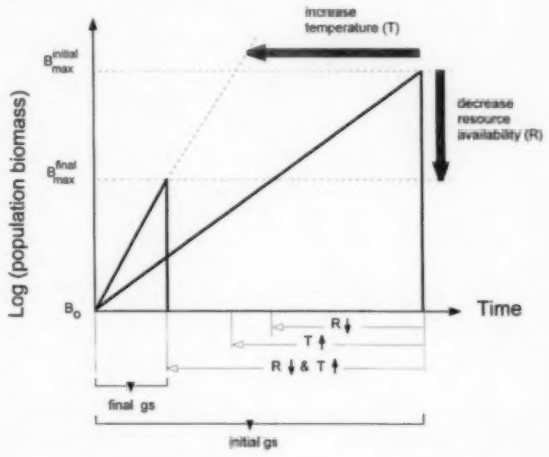
This view suggests a very simple model to illustrate how the interactive effects of temperature and food resource availability (R) may shape the annual production cycle of a particular population. If we assume that (*i*) biomass carrying capacity (B_{max}) for the population is proportional to food resource availability (i.e., $B_{max} \propto R$), (*ii*) population biomass at the start of the annual production cycle (B_0) is small relative to B_{max} and is unrelated to either temperature or resource availability, (*iii*) weight-specific growth follows a simple Van't Hoff function of temperature (i.e., eq. 1) for $B < B_{max}$, and (*iv*) the length of the production cycle (gs) is set by the time required to reach B_{max}, then gs is roughly proportional to $(\log_e[R]/\exp[\beta \cdot T_{gs}])$ and thus both increases in temperature and decreases in food resource availability can act separately, or together, to shorten the annual cycle. We can include the effect of adult body size in this model by making B_{max} proportional to $R \cdot M_{ad}^c$, where $c > 0$. The new M_{ad}^c term captures the hypothesis (Damuth 1981; Lawton 1989) that a fixed level of food resources will support a greater biomass density of larger animals.

The extended model predicts that gs should be positively related to chl and M_{ad} and negatively related to T_{gs}. Further analysis of the observed variation in gs confirmed these predictions. After removing the biased gs values of the long gs group plus one other atypical point from our growing season data set, we obtained the following model ($N = 62$, $R_{adj}^2 = 0.45$, all terms highly significant: $p < 0.004$):

$$(15)\quad \log_{10}(P) = \alpha_{taxon} - 0.020 \cdot T_{gs} + 0.133 \cdot \log_{10}(\text{chl}) + 0.047 \cdot \log_{10}(M_{ad})$$

where α_{taxon} equals 2.37 for Chydoroidea and 2.50 for the other taxonomic groups.

Fig. 4. Schematic plot of seasonal biomass growth illustrating how an increase in temperature and a decrease in resource availability will interact to produce a decrease in the length of the growing season. We assume that biomass growth is exponential and temperature dependent before reaching carrying capacity (B_{max}), carrying capacity varies with resource availability, and when carrying capacity is reached, biomass growth ceases and mortality rapidly depletes biomass.



Our results suggest that the annual production cycle for at least some freshwater zooplankton populations is driven by both temperature and food availability. The interactive effects of these two factors (as summarized in Fig. 4) may be such that a simple increase in temperature, unaccompanied by changes in food availability, would produce a decrease in the length of the annual production cycle, with little change in the overall amount of zooplankton biomass produced and thus made available for transfer up the trophic chain to fish and ultimately to humans. This suggests that predictions of increased fish production from climate change (e.g., Hill and Magnuson 1990) may not be realized in some lake systems.

Acknowledgments

We would like to thank H. Cyr, D. Jackson, J. Stockwell, J. Wong, and an anonymous reviewer for their comments on earlier versions of this manuscript. We are particularly grateful to John Roff for his extensive and perceptive critique. Thanks to the COMPMECH Group at Oak Ridge for getting us interested in the issue. Financial support for this work was provided by the Ontario Ministry of Natural Resources.

References

Adalsteinsson, H. 1979. Zooplankton and its relation to available food in Lake Myvatn. Oikos, **32**: 162–194.

Alimov, A.F., Bouillion, V.V., Finogenova, N.P., Ivanova, M.B., Kuzmitskaya, N.K., Nikulina, V.N., Ozeretskovskaya, N.G., and Zharowa, T.V. 1972. Biological productivity of Lakes Krivoe and Krugloe. *In* Productivity problems in freshwaters. *Edited by* Z. Kajak and A. Hillbricht-Ilkowska. PWN Polish Scientific Publishers, Warszawa-Krakow, Poland. pp. 39–57.

© 1997 NRC Canada

Andrew, T.E. 1983. The estimation of secondary production in a natural population of *Daphnia hyalina* (Leydig) using alternative methods of computation. Hydrobiologia, **107**: 3–18.

Andrew, T.E., and Fitzsimons, A.G. 1992. Seasonality, population dynamics and production of planktonic rotifers in Lough Neagh, Northern Ireland. Hydrobiologia, **246**: 147–164.

Andronikova, I.N., Drabkova, V.G., Kuzmenko, K.N., Michailova, N., and Stravinskaya, E.A. 1970. Biological productivity of the main communities of the Red Lake. *In* Productivity problems in freshwaters. *Edited by* Z. Kajak and A. Hillbricht-Ilkowska. PWN Polish Scientific Publishers, Warszawa-Krakow, Poland. pp. 57–73.

Babitskiy, V.A. 1970. Biology and production of *Eurycercus lamellatus* (O.F.M.) along the shores of Lake Naroch. Hydrobiol. J. **6**: 26–32.

Babitskiy, V.A. 1988. Copepodites of *Mesocyclops leuckarti* in plankton and benthos of a mesotropic lake. Hydrobiol. J. **24**: 11–16.

Balcer, M.D., Korda, N.L., and Dodson, S.I. 1984. Zooplankton of the Great Lakes. The University of Wisconsin Press, Madison, Wis.

Ban, S. 1994. Effect of temperature and food concentration on post-embryonic development, egg production and adult body size of calanoid copepod *Eurytemora affinis*. J. Plankton Res. **16**: 721–735.

Banse, K. 1982. Cell volumes, maximal growth rates of unicellular algae and ciliates, and the role of ciliates in the marine pelagial. Limnol. Oceanogr. **27**: 1059–1071.

Banse, K., and Mosher, S. 1980. Adult body mass and annual production/biomass relationships of field populations. Ecol. Monogr. **50**: 355–379.

Blackburn, T.M., Brown, V.K., Doube, B.M., Greenwood, J.J.D., Lawton, J.H., and Stork, N.E. 1993. The relationship between abundance and body size in natural animal assemblages. J. Am. Ecol. **62**: 519–528.

Borgmann, U., Shear, H., and Moore, J. 1984. Zooplankton and potential fish production in Lake Ontario. Can. J. Fish. Aquat. Sci. **41**: 1301–1309.

Borsheim, K.Y., Andersen, S., Johnsen, G.H., Kambestad, E.O., and Norland, S. 1988. Primary and bacterial production compared to growth and food requirements of *Daphnia longispina* in Lake Kvernavatnet, West Norway. J. Plankton Res. **10**: 921–939.

Bosselman, S. 1975. Production of *Eudiaptomus graciloides* in Lake Esrom, 1970. Arch. Hydrobiol. **76**: 43–64.

Bosselman, S. 1979a. Population dynamics of *Keratella cochlearis* in Lake Esrom. Arch. Hydrobiol. **87**: 152–165.

Bosselman, S. 1979b. Production of *Keratella cochlearis* in Lake Esrom. Arch. Hydrobiol. **87**: 304–313.

Brylinsky, M. 1980. Estimating the productivity of lakes and reservoirs. *In* The functioning of freshwater ecosystems. *Edited by* E.D. LeCren and R.H. Lowe-McConnell. International Biological Programme No. 22. Cambridge Unviersity Press, New York. pp. 411–455.

Burgis, M.J. 1971. The ecology and production of copepods, particularly *Thermocyclops hyalinus* in the tropical Lake George, Uganda. Freshwater Biol. **1**: 169–192.

Burgis, M.J. 1974. Revised estimates for the biomass and production of zooplankton in Lake George, Uganda. Freshwater Biol. **4**: 535–541.

Burgis, M.J., and Walker, A.F. 1972. A preliminary comparison of the zooplankton in a tropical and a temperate lake (Lake George, Uganda, and Loch Leven, Scotland). Int. Ver. Theor. Angew. Limnol. Verh. **18**: 647–655.

Burns, C.W. 1979. Population dynamics and production of *Boeckella dilatata* (Copepoda: Calanoidea) in Lake Hayes, New Zealand. Arch. Hydrobiol. Suppl. **54**: 409–465.

Cajander, V.-R. 1983. Production of planktonic Rotatoria in Ormajärvi, an eutrophicated lake in southern Finland. Hydrobiologia, **104**: 329–333.

Carpenter, S.J., and Kitchell, J.F. 1993. Trophic cascade in lakes. Cambridge University Press, Cambridge, U.K.

Chow-Fraser, P., Trew, D.O., Finlay, D., and Stainton, M. 1994. A test of hypotheses to explain the sigmoidal relationship between total phosphorus and chlorophyll *a* concentrations in Canadian lakes. Can. J. Fish. Aquat. Sci. **51**: 2052–2065.

Clegg, J. 1963. Pond and streamlife of Europe. Blandford Press, London. (English translation of G. Mandahl-Barth).

Crisp, D.J. 1984. Energy flow measurements. *In* Methods for the study of marine benthos. *Edited by* N.A. Holme and A.D. McIntyre. Blackwell, Oxford. pp. 284–372.

Culver, D.A., Boucherle, M.M., Bean, D.J., and Fletcher, J.W. 1985. Biomass of freshwater crustacean zooplankton from length–weight regressions. Can. J. Fish. Aquat. Sci. **42**: 1380–1390.

Cummins, K.W., and Wuycheck, J.C. 1971. Caloric equivalents for investigations in ecological energetics. Int. Ver. Theor. Angew. Limnol. Verh. **18**.

Cyr, H., and Pace, M.L. 1993. Allometric theory: extrapolations from individuals to communities. Ecology, **74**:1234–1245.

Damuth, J. 1981. Population density and body size in mammals. Nature (Lond.), **290**: 699–700.

De Bernardi, R. 1984. Methods for the estimation of zooplankton abundance. *In* A manual on methods for the assessment of secondary productivity in fresh waters. 2nd ed. *Edited by* J.A. Downing and F.H. Rigler. Blackwell Scientific Publishers, Oxford. pp. 59–86.

Dickie, L.M., Kerr, S.R., and Boudreau, P.R. 1987. Size-dependent processes underlying regularities in ecosystem structure. Ecol. Monogr. **57**: 233–250.

Downing, J.A. 1984. Assessment of secondary production: the first step. *In* A manual on methods for the assessment of secondary productivity in fresh waters. 2nd. ed. *Edited by* J.A. Downing and F.H. Rigler. Blackwell Scientific Publishers, Oxford. pp. 1–18.

Edmondson, W.T. 1959. Freshwater biology. John Wiley and Sons, Inc., London.

Edmondson, W.T. 1974. Secondary production. Mitt. Int. Ver. Limnol. **20**: 229–272.

Forsyth, D.J., and James, M.R. 1991. Population dynamics and production of zooplankton in eutrophic Lake Okaro, North Island, New Zealand. Arch. Hydrobiol. **120**: 287–314.

Geller, W. 1987. On estimating the age and the development time of *Daphnia* as a function of body size and temperature. J. Plankton Res. **9**:1225–1230.

Geller, W. 1989. The energy budget of two sympatric *Daphnia* species in Lake Constance: productivity and energy residence times. Oecologia, **78**: 242–250.

George, D.G. 1976. Life cycle and production of *Cyclops vicinus* in a shallow eutropic reservoir. Oikos, **27**: 101–110.

Gillespie, D.M., and Benke, A.C. 1979. Methods of calculating cohort production from field data — some relationships. Limnol. Oceanogr. **24**: 171–176.

Glazier, D., and Calow, P. 1992. Energy allocation rules in *Daphnia magna:* clonal and age differences in the effect of food limitation. Oecolgia, **90**: 540–549.

Gophen, M. 1978. The productivity of *Mesocyclops leuckarti* in Lake Kinneret, Israel. Hydrobiologia, **60**: 17–22.

Gophen, M. 1988. Changes of Copepoda populations in Lake Kinneret during 1969–1985. Hydrobiologia, **167/168**: 375–379.

Gophen, M., Serruya, S., and Spataru, P. 1990. Zooplankton community changes in Lake Kinneret (Israel) during 1969–1985. Hydrobiologia, **191**: 39–46.

© 1997 NRC Canada

Green, J.D. 1976. Population dynamics and production of the calanoid copepod *Calamoecia lucasi* in a northern New Zealand lake. Arch. Hydrobiol. Suppl. **50**: 313–400.

Hanazato, T., and Yasuno, M. 1985. Population dynamics and production of cladoceran zooplankton in the highly eutropic Lake Kasumigaura. Hydrobiologia, **124**: 13–23.

Hanson, J.M., and Peters, R.H. 1984. Empirical prediction of crustacean zooplankton biomass and profundal macrobenthos biomass in lakes. Can. J. Fish. Aquat. Sci. **41**: 439–445.

Hart, R.C. 1990. Copepod post-embryonic durations: pattern, conformity and predictability. The realities of isochronal and equiproportional development, and trends in the copepodid–naupliar duration ratio. Hydrobiologia, **206**: 175–206.

Hart, R.C. 1991. Food and suspended sediment influence on the naupliar and copepodid durations of freshwater copepods: comparative stuides on *Tropodiaptomus* and *Metadiaptomus*. J. Plankton Res. **13**: 645–660.

Hedges, L.V., and Olkin, I. 1985. Statistical methods for meta-analysis. Academic Press, New York.

Herzig, A., Anderson, R.S., and Mayhood, days.W. 1980. Production and population dynamics of *Leptodiaptomus sicilis* in a mountain lake in Alberta, Canada. Holarct. Ecol. **3**: 50–63.

Hill, D.K., and Magnuson, J.J. 1990. Potential effects of global climate warming on the growth and prey consumption of Great Lakes fish. Trans. Am. Fish. Soc. **119**: 265–275.

Hillbricht-Ilkowska, A., and Weglenska, T. 1970. Some relations between production and zooplankton structure of two lakes of varying trophy. Pol. Arch. Hydrobiol. **17**: 233–240.

Humphreys, W.F. 1979. Production and respiration in animal populations. J. Anim. Ecol. **48**: 427–453.

Humphreys, W.F. 1981. Towards a simple index based on live-weight and biomass to predict assimilation in animal populations. J. Anim. Ecol. **50**: 543–561.

Huntley, M.E., and Lopez, M.D.G. 1992. Temperature-dependent production of marine copepods: a global synthesis. Am. Nat. **140**: 201–242.

Hutchinson, G.E. 1967. A treatise on limnology. Vol. II. Introduction to lake biology and the limnoplankton. John Wiley and Sons, Inc., New York.

Jackson, D.A., Harvey, H.H., and Somers, K.M. 1991. Ratios in aquatic sciences: statistical shortcomings with mean depth and the morphoedaphic index. Can. J. Fish. Aquat. Sci. **47**: 1788–1795.

Jamieson, C., and Burns, C. 1988. The effects of temperature and food on copepodite development, growth and reproduction in three species of *Boeckella* (Copepoda: Calanoida). Hydrobiologia, **164**: 235–257.

Jones, H.R., Lack, T.J., and Jones, C.S. 1979. Population dynamics and production of *Daphnia hyalina* var. *lacustris* in Farmoor I, a shallow eutrophic reservoir. J. Plankton Res. **1**: 45–65.

King, C.R., and Greenwood J.G. 1992*a*. The seasonal population changes and carbon budget of the calanoid copepod *Boeckella minuta* Sars in a newly formed sub-tropical reservoir. J. Plankton Res. **14**: 329–342.

King, C.R., and Greenwood, J.G. 1992*b*. Productivity and carbon budget of a natural population of *Daphnia lumholtzi* Sars. Hydrobiologia, **231**: 197–207.

Larsson, P. 1978. The life cycle dynamics and production of zooplankton in Ovre Heimdalsvatn. Holarct. Ecol. **1**: 162-218.

Lawton, J.H. 1989. What is the relationship between population density and body size in animals? Oikos, **55**: 429–434.

Lévêque, C., and Saint-Jean, L. 1983. III. The balance of a lacustrine ecosystem during "normal" Chad and a period of drought. Secondary production (zooplankton and benthos). *In* Lake Chad: ecology and productivity of a shallow tropical lake ecosystem. *Edited by* J.-P. Carmouze, J.R. Durand, and C. Leveque. W. Junk Publishers, The Hague, The Netherlands. Monogr. Biol. **53**: 385–424.

Lynch, M. 1980. The evolution of cladoceran life histories. Q. Rev. Biol. **55**: 23–42.

Lynch, M. 1989. The life history consequences of resource depression in *Daphnia pulex*. Ecology, **70**: 246–256.

MacCallum, M.E. 1971. Distribution, population dynamics and production of *Keratella cochlearis* in an Arctic Lake. M.Sc. thesis, University of Toronto, Toronto, Ont.

Makarewicz, J.A., and Likens, G.L. 1979. Structure and function of the zooplankton community of Mirror Lake, New Hampshire. Ecol. Monogr. **49**: 109–127.

Mason, C.F., and Abdul-Hussein, M.M. 1991. Population dynamics and production of *Daphnia hyalina* and *Bosmina longirostris* in a shallow, eutrophic reservoir. Freshwater Biol. **25**: 243–260.

Matsumura-Tundisi, T., Rietzler, A.C., and Tundisi, J.G. 1989. Biomass (dry weight and carbon content) of plankton Crustacea from Broa reservoir (Sao Carlos, S.P.-Brazil) and its fluctuation across one year. Hydrobiologia, **179**: 229–236.

Mavuti, K.M. 1990. Ecology and role of zooplankton in the fishery of Lake Naivasha. Hydrobiologia, **208**: 131–140.

Mavuti, K.M. 1994. Durations of development and production estimates by two crustacean zooplankton species, *Thermocyclops oblongatus* Sars (Copepoda) and *Diaphanosoma excisum* Sars (Cladocera), in Lake Naivasha, Kenya. Hydrobiologia, **272**: 185–200.

Mazumder, A. 1994. Phosphorus–chlorophyll relationships under contrasting herbivory and thermal stratification: predictions and patterns. Can. J. Fish. Aquat. Sci. **51**: 390–400.

McCauley, E. 1984. The estimation of the abundance and biomass of zooplankton in samples. *In* A manual on methods for the assessment of secondary productivity in fresh waters. *Edited by* J.A. Downing and F.H. Rigler. Blackwell Scientific Publications, Oxford. pp. 228–265.

McCauley, E., and Kalff, J. 1981. Empirical relationships between phytoplankton and zooplankton biomass in lakes. Can. J. Fish. Aquat. Sci. **38**: 458–463.

McCauley, E., Murdoch, W.W., and Nisbett, R.M. 1990*a*. Growth, reproduction and mortality of *Daphnia pulex* Leydig: life at low food. Funct. Ecol. **4**: 505–514.

McCauley, E., Murdoch, W.W., Nisbett, R.M., and Gurney, W. 1990*b*. The physiological ecology of *Daphnia*: development of a model of growth and reproduction. Ecology, **71**: 703–715.

McQueen, D.J., Johannes, M.R.S., Post, J.R., Stewart, T.J., and Lean, days.R.S. 1989. Bottom-up and top-down impacts on freshwater pelagic community structure. Ecol. Monogr. **59**: 289–309.

Mengestou, S., and Fernando, C.H. 1991*a*. Biomass and production of the major dominant crustacean zooplankton in a tropical Rift Valley lake, Awasa, Ethiopia. J. Plankton Res. **13**: 831–851.

Mengestou, S., and Fernando, C.H. 1991*b*. Seasonality and abundance of some dominant crustacean zooplankton in Lake Awasa, a tropical Rift Valley lake in Ethiopia. Hydrobiologia, **209**: 203–214.

Morin, A., and Bourassa, N. 1992. Modèles empiriques de la production annuelle et du rapport *P/B* d'invertébrés benthiques d'eau courante. Can. J. Fish. Aquat. Sci. **49**: 532–539.

Pace, M.L. 1984. Zooplankton community structure, but not biomass, influences the phosphorus – chlorophyll *a* relationship. Can. J. Fish. Aquat. Sci. **41**: 1089–1096.

Paloheimo, J.E., Crabtree, S.J., and Taylor, W.D. 1982. Growth model of *Daphnia*. Can. J. Fish. Aquat. Sci. **39**: 598–606.

Pennak, R.W. 1989. Freshwater invertebrates of the United States. John Wiley and Sons, Inc., New York.

Peters, R.H. 1983. The ecological implications of body size. Cambridge University Press, Cambridge, U.K.

© 1997 NRC Canada

Peters, R.H., and Wassenberg, K. 1983. The effect of body size on animal abundance. Oecologia (Berlin), **60**: 89–96.

Plante, C., and Downing, J.A. 1989. Production of freshwater invertebrate populations in lakes. Can. J. Fish. Aquat. Sci. **46**: 1489–1498.

Rey, J., and Capblancq, J. 1975. Dynamique des populations et production de zooplankton du lac de Port-Bielh (Pyrenees centrales). Ann. Limnol. **11**: 1–45.

Rigler, F.H., and Downing, J.A. 1984. The calculation of secondary productivity. *In* A manual on methods for the assessment of secondary productivity in fresh waters. 2nd ed. *Edited by* J.A. Downing and F.H. Rigler. Blackwell Scientific Publishers, Oxford. pp. 19–58.

Rigler, F.H., MacCallum, M.E., and Roff, J.C. 1974. Production of zooplankton in Char Lake. J. Fish. Res. Board Can. **31**: 637–646.

Rocha, O., and Matsumura-Tundisi, T. 1984. Biomass and production of *Argyrodiaptomus durcatusa furcatusa* tropical calanoid copepod in Broa Reservoir, Southern Brazil. Hydrobiologia, **113**: 307–312.

Santer, B., and van den Bosch, F. 1994. Herbivorous nutrition of *Cyclops vicinus*. The effect of pure algal diet on feeding, development and reproduction and life cycle. J. Plankton Res. **12**: 549–572.

Schindler, D.W. 1972. Production of phytoplankton and zooplankton in Canadian Shield lakes. *In* Productivity problems in freshwaters. *Edited by* J. Kajak and A. Hillbricht-Ilkowska. PWN Polish Scientific Publishers, Warszawa-Krakow, Poland. pp. 311–333.

Schmidt-Nielsen, K. 1984. Scaling: why is animal size so important? Cambridge University Press, Cambridge, U.K.

Sokal, R.R., and Rohlf, F.J. 1981. Biometry. W.H. Freeman, San Francisco, Calif.

Sommer, U. 1985. Seasonal succession of phytoplankton in Lake Constance. BioScience, **35**: 351–357.

Sommer, U. 1989. Maximal growth rates of Antarctic phytoplankton: only weak dependence on cell size. Limnol. Oceanogr. **34**: 1109–1112.

Straskraba, M. 1980. The effects of physical variables on models. *In* The functioning of freshwater ecosystems. *Edited by* E.D. LeCren and R.H. Lowe-McConnell. International Biological Programme No. 22, Cambridge University Press, Cambridge. pp. 13–85.

Strayer, D.L. 1994. Body size and abundance of benthic animals in Mirror Lake, New Hampshire. Freshwater Biol. **32**: 83–90.

Sutcliffe, W.H., Loucks, R.H., and Drinkwater, K.F. 1976. Coastal circulation and physical oceanography of the Scotian Shelf and the Gulf of Maine. J. Fish. Res. Board Can. **33**: 98–115.

Thorp, J.H., and Covich, A.P. 1991. Ecology and classification of North American freshwater invertebrates. Academic Press, Inc., San Diego, Calif.

Trippel, E.A. 1991. Potential effects of global warming on whitefish in Lake Constance, Germany. Ambio, **20**: 226–231.

Tumbiolo, M.L., and Downing, J.A. 1994. An empirical model for the prediction of secondary production in marine benthic invertebrate populations. Mar. Ecol. Prog. Ser. **114**: 165–174.

van den Bosch, F., and Gabriel, W. 1994. A model of growth and development in copepods. Limnol. Oceanogr. **39**: 1528–1542.

Vareschi, E. 1982. The ecology of Lake Nakuru (Kenya). III. Abiotic factors and primary production. Oecologia, **55**: 81–101.

Vareschi, E., and Jacobs, J. 1984. The ecology of Lake Nakuru (Kenya). V. Production and consumption of consumer organisms. Oecologia, **61**: 83–98.

Vareschi, E., and Jacobs, J. 1985. The ecology of Lake Nakuru. VI. Synopsis of production and energy flow. Oecologia, **65**: 412–424.

Vareschi, E., and Vareschi, A. 1984. The ecology of Lake Nakuru (Kenya). IV. Biomass and distribution of consumer organisms. Oecologia, **61**: 70-82.

Vijverberg, J., and Frank, Th.H. 1976. The chemical composition and energy contents of copepods and cladocerans in relation to their size. Freshwater Biol. **6**: 333–345.

Vijverberg, J.V., and Richter, A.F. 1982*a*. Population dynamics and production of *Daphnia hyalina* Leidig and *Daphnia cucullata* Sars in Tjeukemeer. Hydrobiologia, **95**: 235–259.

Vijverberg, J.V., and Richter, A.F. 1982*b*. Population dynamics and production of *Acanthocyclops robustus* (Sars) and *Mesocyclops leuckarti* (Clauss) in Tjeukemeer. Hydrobiologia, **95**: 261–274.

Waters, T.F. 1977. Secondary production in inland waters. Adv. Ecol. Res. **10**: 91–164.

Wernstedt, F.L. 1972. World climatic data. Climatic Data Press, Lemont, Pa.

Winberg, G.G., Alimov, A.F., Bouillion, V.V., Ivanova, M.B, Korobtzova, E.V., Kuzmitzkaya, N.K., Nikulina, V.N., Finogenova, N.P., and Fursenko, M.V. 1973. Biological productivity of two sub-arctic lakes. Freshwater Biol. **3**: 177–197.

© 1997 NRC Canada

Effects of navigation dams, tributaries, and littoral zones on phytoplankton communities in the Ohio River[1]

John D. Wehr and James H. Thorp

Abstract: Phytoplankton and water chemistry variables were sampled monthly along a 361-km stretch of the Ohio River. We compared (*i*) regions from Ohio through Kentucky, (*ii*) sites above, below, and within tributaries, (*iii*) nearshore and channel habitats, and (*iv*) sites away from and near navigation dams. Densities of suspended cells were lowest during colder, high-flow periods (31 000 cells/mL) and greatest during warmer, low-flow summer (104 000 cells/mL). Of 134 taxa identified, >60% were diatoms, comprising 6% of cell numbers and 28% of total biovolume. Summer assemblages were dominated by colonial *Aphanothece saxicola*, *Merismopedia punctata*, *Microcystis aeruginosa*, and unicellular *Synechococcus* species. Diatoms *Melosira jurgensii* and *Melosira distans* were abundant in summer and *Cyclotella* spp. in spring. Concentrations of dissolved inorganic phosphorus (≈40 μg/L) and dissolved inorganic nitrogen (≈1100 μg/L) were high; neither was positively correlated with any phytoplankton variable. Interpool declines in picoplankton correlated with greater copepod nauplii densities. Navigation dams significantly reduced current velocity and densities of benthic forms; dissolved inorganic phosphorus levels were significantly greater. Near-dam sites had increased biomass of larger phytoplankton (>2 μm) in winter, but lower densities during summer. Multiple regression identified temperature (+) and current velocity (−) as the main factors affecting phytoplankton densities in this large river.

Résumé : Le phytoplancton a été échantillonné et les caractéristiques chimiques de l'eau analysées tous les mois le long d'un segment de 361 km de la rivière Ohio. Nous avons comparé (*i*) les régions de l'Ohio au Kentucky, (*ii*) des sites en amont, en aval et à l'intérieur des tributaires, (*iii*) les habitats près des rives et dans les chenaux et (*iv*) les sites éloignés et proches des barrages pour la navigation. La densité des cellules en suspension était plus faible durant les périodes plus froides et de hautes eaux (31 000 cellules/mL) et plus élevée durant les périodes plus chaudes à basses eaux de l'été (104 000 cellules/mL). Des 134 taxons identifiés, >60% étaient des diatomées, constituant 6% du nombre de cellules et 28% du biovolume total. Les assemblages d'été étaient dominés par *Aphanothece saxicola*, *Merismopedia punctata* et *Microcystis aeruginosa*, organismes vivant en colonies, et par les espèces de *Synechococcus*, organismes unicellulaires. Les diatomées *Melosira jurgensii* et *Melosira distans* étaient abondantes durant l'été, et les espèces *Cyclotella* au printemps. Les concentrations de phosphore inorganique dissous (≈40 μg/L) et d'azote inorganique dissous (≈1100 μg/L) étaient élevées; ni l'une ni l'autre n'était corrélée positivement avec une quelconque variable liée au phytoplancton. Les diminutions interfosses du picoplancton étaient en corrélation avec les densités élevées de nauplii de copépodes. Les barrages de navigation ont réduit substantiellement la vitesse du courant et la densité des organismes benthiques; les concentrations de phosphore inorganique dissous étaient nettement plus élevées. Les sites près des barrages présentaient une biomasse accrue de phytoplancton de plus grande taille (>2 μm) en hiver, mais des densités plus faibles en été. La régression multiple a révélé que la température (+) et la vitesse du courant (−) étaient les principaux facteurs influant sur la densité du phytoplancton dans cette grande rivière.
[Traduit par la Rédaction]

Introduction

The large majority of ecological studies on autotrophic organisms of flowing waters has concentrated on streams and small rivers (Hynes 1989; Thorp et al. 1994). Of the studies that concern larger rivers, many have focused on attached (periphyton) algal communities, such as in the Columbia and the Mississippi rivers (Cushing 1967; Luttenton et al. 1986). Such studies are needed because large rivers can support substantial benthic primary production (Wetzel 1975). Unfortunately, there is a persistent misunderstanding that the phytoplankton of large rivers is sparse and consists primarily of detached benthic forms suspended in the water column (Whitton 1975; Vannote et al. 1980). This misinformation is particularly surprising given the long history of floristic accounts of phytoplankton in rivers of Europe and North America. One of the earliest detailed studies of river algae was by Zacharias (1898), who characterized the phytoplankton in several large rivers in Germany and suggested that densities were inversely related to flow conditions. In North America, Kofoid (1903) reported on the planktonic organisms of the Illinois River between 1894 and 1899, observing blooms of cyanobacteria

Received March 13, 1996. Accepted July 15, 1996.
J13352

J.D. Wehr.[2] Louis Calder Center and Department of Biology, Fordham University, P.O. Box K, Armonk, NY 10504, U.S.A.
J.H. Thorp. Large Rivers Program, Biology Department, University of Louisville, Louisville, KY 40292, U.S.A.

[1] Contribution No. 142 to the Louis Calder Center.
[2] Author to whom all correspondence should be addressed.
e-mail: wehr@murray.fordham.edu

© 1997 NRC Canada

during the summer and diatom maxima in autumn and winter. In the intervening years, many studies described the potamoplankton of large rivers, but there continued to be a debate concerning whether a true phytoplankton community exists in rivers, or whether the assemblage consists primarily of resuspended cells from tributaries and lakes within the associated floodplain (see reviews by Hynes 1970; Whitton 1975).

More recent research on flowing waters has centered around the concepts proposed in the River Continuum Concept (RCC: Vannote et al. 1980). Among many ideas, the RCC predicts that in large-order (greater than sixth) rivers, phytoplankton production is light limited and that self-sustaining algal populations are unimportant to ecosystem metabolism because most large rivers are deep and turbid. This view has recently been challenged by authors who contend that floodplain inputs, littoral zone communities, and in situ production may be considerable in large rivers (Junk et al. 1989; Thorp and Delong 1994). Another reason for a change of view is that most large rivers of the world are regulated to varying degrees by channelization (bank stabilization) and high or low (navigation) dams. The effects of these water-control measures are many, but a common feature is the creation of small impoundments or pools in which current velocity is reduced (see discussions by Lack 1971; Søballe and Kimmel 1987; Thorp and Delong 1994; Thorp et al. 1994). These modifications also reduce effects of turbidity and downstream loss, factors that are predicted by the RCC to limit phytoplankton production in large unregulated rivers (Vannote et al. 1980).

To understand the role of resident phytoplankton communities in large regulated rivers, studies first need to identify the primary sources of these algae, as well as how communities respond to factors that characterize most large rivers of today. In connection with broader studies on the Ohio River ecosystem, we describe the first step in characterizing these influences on the phytoplankton communities over an annual cycle. Specifically, we examine the impacts of littoral communities, tributary rivers, and navigation dams on the development of phytoplankton assemblages. We also measured a series of physical and chemical variables that also may be affected by hydrological and anthropogenic features that are common to many large rivers worldwide.

Methods

Study sites and sampling

This study was conducted in parallel with a previously published account on the zooplankton of the Ohio River (Thorp et al. 1994) that provides details of the hydrology and other physical features of the Ohio and a full description of the size and location of study sites. Overall, our study spans a 361 km long stretch of the river beginning downstream of Cincinnati, Ohio, to about 60 km upstream of Evansville, Ind. The study was based on 12 monthly samples from February 1991 through January 1992 from three large reaches of the Ohio River (termed pools locally): (*i*) Markland pool (Ma; 155 km long), an upper constricted channel; (*ii*) McAlpine pool (Mc; 119 km), a midriver constricted section; and (*iii*) Cannelton pool (Ca; 185 km), which lies in a broader floodplain region downstream of Louisville, Ky. By design, each study pool was selected to include one major tributary (the Miami, Kentucky, and Salt rivers, respectively) in order to examine the influence of floodplain inputs and tributaries on the plankton and nutrient pools of the main river (see Thorp et al. 1994 for further details). Within each 100+ km pool, a 2 × 4 sampling design included two habitats, pelagic (Channel) and littoral (Bank), and four sites, above tributary (ATR), in tributary (ITR), below tributary (BTR), and just above the navigation dam (LOW). All samples were spatially replicated five times. This resulted in a total of 3 pools × 4 sites × 2 habitats × 5 replicates = 120 samples collected each month.

On each sample date, five replicate phytoplankton samples were collected at a depth of 0.5–1.0 m using 20-L plastic buckets at each pool–site from a boat. Unconcentrated subsamples were placed in 20-mL plastic scintillation vials and preserved with Lugol's iodine until laboratory analysis. Physical measurements were made in situ as follows. Current velocity in the pelagic and littoral areas was measured with a Marsh McBirney velocity meter (model 201-D), and water temperatures were recorded with a YSI temperature probe. Water samples were retrieved for turbidity analysis from the main river sites and measured in NTU units with a Hach turbidimeter. Water samples for chemical analysis were filtered in line (Whatman GF/F) and frozen (–15°C) for later processing.

Laboratory analyses

Subsamples (1–5 mL) of phytoplankton samples from all seasons and sites were observed to identify most taxa and establish a species list for later counting. This list represented >80% of all taxa encountered during the entire study. For enumeration, 10-mL aliquots of Lugol's preserved samples were placed in sedimentation chambers and allowed to settle for a minimum of 12 h. Identification and counting procedures followed the standard inverted microscope method (Utermöhl 1958), using a Nikon Diaphot microscope. A minimum of 10 grids and 1000 cells were enumerated from each sample.

Water chemistry samples were analyzed for concentrations of dissolved inorganic (=soluble-reactive) P (DIP) using antimony–ascorbate–molybdate (American Public Health Association 1985; Bran+Luebbe Analyzing Technologies 1986*a*), NH_4^+-N using phenol–hypochlorite, and NO_3^- (after reduction to NO_2^- in a Cd–Cu column) via reaction with sulfanilamide–NNED (American Publich Health Association 1985; Bran+Luebbe Analyzing Technologies 1986*b*, 1987*a*). Soluble-reactive Si (as SiO_2) was measured via the molybdosilicate method (American Public Health Association 1985; Bran+Luebbe Analyzing Technologies 1987*b*), and SO_4-S was analyzed using the $BaCl_2$ method (American Publich Health Assocation 1985; Bran+Luebbe Analyzing Technologies 1989). The above procedures were modified for automated analysis and run on a Traacs 800 automated analyzer (Bran+Luebbe Inc., Buffalo Grove, Ill.). Ca and Mg concentrations were measured using flame atomic absorption spectroscopy (Perkin-Elmer model 1100B).

Data analysis and statistics

The primary aim of our analysis was to test whether "structural" features of the Ohio River (regional pools, dams, tributaries, and littoral–pelagic habitats) significantly affected phytoplankton densities and water chemistry in the river. The questions posed in our objectives were addressed in each of four separate river seasons that we identified previously, based on temperature and flow conditions (Thorp et al. 1994). Data analysis was preceded by classifying the 134 algal and cyanobacterial taxa into six ecological (total: total >2 μm; planktonic forms: plankton >2 μm; benthic forms: picoplankton) and six taxonomic (cyanobacteria, cyanobacteria >2 μm, Chlorophyta, Chrysophyta, Cryptophyta + Dinophyta, Bacillariophyta) categories. Species assigned to the benthic category were based on designations of Lowe (1974) and Whitton (1975). These 12 groups served as the dependent variables for all hypothesis tests.

Prior to analyses, all data were checked for assumptions of normality and homogeneity of variances. Of 24 variables tested, 20 were strongly skewed, but a $\log_{10}$ transformation enabled the use of parametric statistical tests. Downstream effects of regional pools were analyzed using analysis of variance (ANOVA), with post hoc analyses using Tukey's honestly significant difference (HSD) among

© 1997 NRC Canada

Table 1. Physical conditions in the main channel of the Ohio River during February 1991 to January 1992.

	Markland pool			McAlpine pool			Cannelton pool		
Variable	ATR	BTR	LOW	ATR	BTR	LOW	ATR	BTR	LOW
Temperature (°C)									
Min.	5.5	5.5	4.5	4.0	4.0	3.0	4.0	4.0	5.9
Mean	15.8	15.8	14.2	15.2	15.0	13.8	14.5	14.7	15.2
Max.	29.0	28.0	26.0	27.0	26.0	25.0	26.5	27.0	27.0
Current (m/s)									
Min.	0.05	0.08	0.09	0.10	0.17	0.14	0.21	0.22	0.17
Mean.	0.53	0.46	0.44	0.77	0.70	0.32	0.60	0.68	0.46
Max.	1.21	1.42	1.30	1.68	1.61	0.68	1.10	1.18	0.85
Turbidity (NTU)									
Min.	4.0	5.5	3.0	2.0	2.5	2.0	2.0	2.5	2.0
Mean	17.9	14.8	17.5	18.8	16.4	12.6	17.2	17.4	12.3
Max.	39.5	41.0	46.0	50.0	40.0	33.0	34.0	35.5	25.5

Note: n = 12 for each pool-site grouping; ATR, above tributary; BTR, below tributary; LOW, at navigation dam.

individual groups (Sokol and Rohlf 1995). Effects of hypothesized factors (navigation dams, tributaries, pelagic versus littoral) were analyzed using a stratified (monthwise) Student t test. Relationships between measured physical and chemical factors and phytoplankton abundances were first examined using (Pearson's) linear correlation. For all hypothesis tests, effects were judged to be significant if the probability of an event having occurred owing to chance (P) was <0.05. A more complete set of analyses examined the combined influences of physical and chemical variables on the phytoplankton community, using stepwise multiple linear regression (MLR). In these, 12 independent variables were tested for their effects on phytoplankton: Ca^{2+}, Mg^{2+}, DIP, NH_4^+-N, NO_3^--N, total dissolved inorganic N (DIN), Si, SO_4-S, the inorganic N:P ratio, current velocity, temperature, and turbidity. Because regression relationships between putative positive factors and phytoplankton abundance may be either positive (=stimulation) or negative (=consumption), we are applying these empirical models mainly as a guide to examining broad patterns in the river. The maximum criterion for inclusion of independent variables into each model was $\alpha = 0.05$, and the minimum for exclusion was $\alpha = 0.10$. Data were complied and analyzed using SYSTAT 5.1 (Wilkinson 1992).

Results

Physical and chemical conditions

Seasonal patterns of physical and chemical conditions described in Thorp et al. (1994) are summarized here to emphasize differences as they affect the phytoplankton communities among pools, sites, and specific months under study (Table 1). Average current velocities varied among sites, but overall were not significantly less near the navigation dams (LOW sites). However, specific patterns within each regional pool did occur. On average, current velocity was reduced near the McAlpine dam (t test: $P < 0.05$), but was not significantly different at either the Markland or Cannelton dam over the year (t tests: $P = 0.78$ and $P = 0.13$, respectively). This decline in current in the lower McAlpine pool did not coincide with lower (nonalgal) turbidity levels here or at any LOW site ($P > 0.05$). Tributary inputs and navigation dams had apparently no significant effects on either variable in the main river. Average, minimum, and maximum temperatures exhibited no consistent or significant pattern along the downriver gradient or with respect to the presence of dams or tributaries.

Chemical conditions varied considerably along the course of the river (Table 2; Fig. 1). DIP concentrations averaged 43 μg P/L riverwide and differed among sites during April, July, and January (ANOVA: $P < 0.05$), but not during October ($P = 0.068$). Tributaries had no significant overall effect on DIP concentrations in the main river, regardless of time of year (t test: $P > 0.05$). DIP concentrations were, however, lower in sites near navigation dams during April and October (t tests: $P = 0.001$ and $P = 0.018$, respectively). Regional (downstream) patterns (Fig. 1) suggest that DIP was relatively constant along the entire river in April and during all months within the upper Markland region of the Ohio (ANOVA: $P > 0.05$). Further downstream, DIP concentrations varied more widely. The most pronounced changes in DIP along the river gradient were observed in the McAlpine region, particularly above and below the McAlpine dam (Mc LOW). DIP concentrations more than doubled in Mc LOW during July and January and decreased by roughly 50% at the same site during October.

DIN concentrations in the Ohio were typically high, with an annual average of 1120 μg N/L, most of which was NO_3-N (Table 2; Fig. 2). DIN levels were similar along the main regions of the river, with the exception of one highly significant site effect during January (ANOVA: $P < 0.001$). Tributaries and navigation dams apparently did not affect DIN levels ($P > 0.05$), with the exception of greater concentrations at the McAlpine Dam during July and January and lower levels of DIN during October (t test: $P < 0.05$). Dissolved Si levels were low (annual average <5 mg/L at all pools and sites) and varied by only a factor of roughly 5 over the annual cycle. None of

© 1997 NRC Canada

Table 2. Chemical variables measured in the main channel of the Ohio River during February 1991 to January 1992.

	Markland pool			McAlpine pool			Cannelton pool		
Variable	ATR	BTR	LOW	ATR	BTR	LOW	ATR	BTR	LOW
DIP (µg P/L)									
Min.	21.3	15.3	22.2	3.8	4.5	15.7	13.0	21.9	21.9
Mean	39.8	39.9	39.6	34.9	43.0	52.1	55.6	48.7	36.8
Max.	71.4	79.8	58.3	81.0	78.3	102.4	100.9	80.1	69.0
NH_4-N (µg N/L)									
Min.	68	23	78	4	12	17	66	3	32
Mean	227	225	163	157	184	153	262	231	199
Max.	737	864	442	433	513	600	643	727	482
NO_3-N (µg N/L)									
Min.	189	214	202	165	167	158	191	263	200
Mean	819	799	886	727	788	919	894	735	716
Max.	1473	1619	1543	1655	1602	2151	1818	1583	1553
DIN (µg/L)									
Min.	286	314	333	303	312	232	634	362	294
Mean	1124	1106	1143	941	1004	1148	1236	1043	985
Max.	1779	1854	1828	1840	1831	2410	2470	1722	1666
N:P ratio									
Min.	11.6	12.6	12.0	11.5	6.6	3.8	9.6	7.1	12.6
Mean	30.4	33.0	30.0	37.1	29.0	28.2	27.0	24.0	28.2
Max.	49.6	63.5	48.5	178.9	129.9	65.9	76.1	57.6	45.5
Si (mg/L)									
Min.	1.9	2.0	2.1	0.7	2.2	1.0	1.9	<0.1	1.1
Mean	4.4	4.6	4.8	3.5	4.1	3.7	4.4	4.1	3.3
Max.	6.3	9.0	10.9	8.1	6.0	6.6	9.5	14.1	6.9
SO_4-S (mg S/L)									
Min.	23.4	11.8	14.1	9.2	3.4	10.0	20.0	5.8	8.0
Mean	69.2	61.1	62.2	54.4	55.3	55.2	55.1	50.6	51.7
Max.	111.8	115.1	116.0	137.2	128.2	127.2	108.8	115.6	114.4
Ca (mg/L)									
Min.	14.4	6.0	13.3	10.0	9.2	8.8	10.2	11.5	6.5
Mean	31.6	29.5	32.4	22.0	30.1	23.1	35.7	33.5	33.2
Max.	44.9	46.4	43.5	38.1	80.4	46.5	53.6	50.8	48.9
Mg (mg/L)									
Min.	5.5	1.6	2.0	1.9	1.4	1.5	3.0	1.7	1.5
Mean	9.0	10.3	8.9	7.9	8.2	8.7	9.4	9.5	8.8
Max.	13.8	25.2	14.5	16.8	15.2	15.9	14.8	16.1	16.1

Note: n = 60 for each; ATR = above tributary, BTR = below tributary, LOW = at navigation dam.

the above data exhibited temporally or spatially consistent downstream patterns, such as increasing eutrophy along the river gradient.

Phytoplankton community

During 1991–1992, we observed a total of 134 algal and cyanobacterial taxa, most of which were recognized to species level (Appendix). Most of the phytoplankton species diversity was contributed by diatoms (Bacillariophyta; 82 taxa, about 60%), although a majority of these taxa were rarely observed or occurred in low densities. The importance of members of the cyanobacteria and Chlorophyta was greatest in warmer months. For example, 14 cyanobacterial and 22 green algal taxa were identified from these two groups in July, but only 11 and 9 were recognized in January. Few representatives of the dinoflagellates, cryptophytes, and chrysophytes were seen during any time of the year. In contrast, diatom species number was lowest in July (30 taxa) and greatest in April (59 taxa).

© 1997 NRC Canada

Fig. 1. Concentrations of DIP (mean ± 1 SE) in the nine stations of the Ohio River measured during four seasons during 1991–1992. n = 10 for each point; broken lines indicate the 95% confidence intervals of the mean concentration on that date.

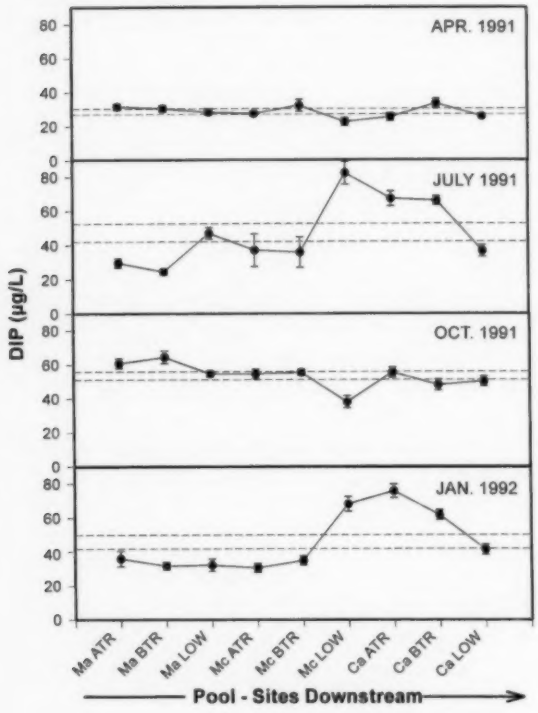
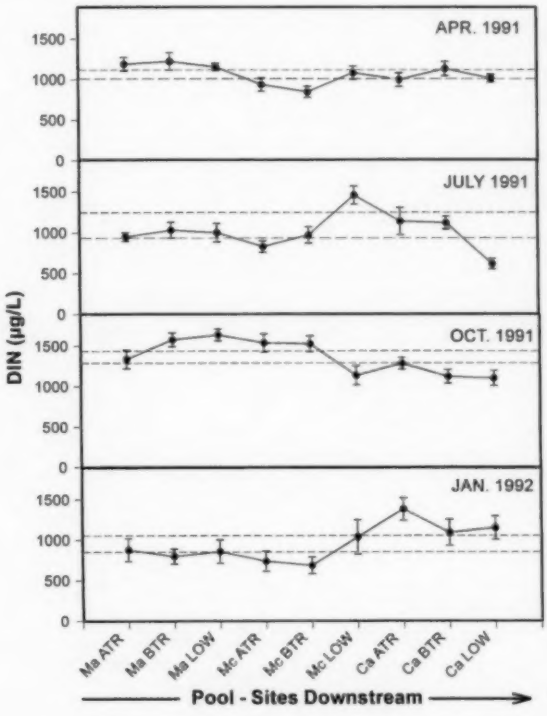


Fig. 2. Concentrations of DIN (=Σ NO_3 + NO_2 + NH_4) (mean ± 1 SE) in the nine stations of the Ohio River measured during four seasons during 1991–1992. n = 10 for each point; broken lines indicate the 95% confidence intervals of the mean concentration on that date.



Abundances of the 10 most important individual taxa (most to species level) also varied according to time of year (Fig. 3). A number of species were encountered in nearly all samples and on most dates. By far the most ubiquitous and abundant taxon was *Synechococcus* sp., a collection of small cyanobacteria <2 µm in size (annual average 62% of cells, 9.9% of total biovolume). Its maximum density was observed in April and May (riverwide average ≈65 000/mL) and minimum in late autumn (October–November average ≈22 000/mL). Whether this category represents one or several species is unknown. The colonial cyanobacteria *Aphanothece saxicola*, *Merismopedia tennuissima*, *Microcystis aeruginosa*, and *Pseudanabaena catanata* were abundant only during warmer months (June– October). The most important diatom species, *Melosira jurgensii* and *Melosira distans*, also were more abundant during the warmer months (June–October). Other important species, including the diatoms *Cyclotella comta*, *Cyclotella meneghiniana*, *Asterionella formosa*, and *Fragilaria crotonensis* (last two not shown), were more abundant during the cooler months of November–January. Other ubiquitous nondiatom taxa, including *Scendesmus quadricauda*, coccoid cells 2–4 µm, and a small unidentified flagellated chrysophyte, tended to be more abundant in spring–summer. A few taxa were sporadically abundant but rare during other times of the year, such as *Gonium pectorale*, *Pseudanabaena catanata*, and *Chroococcus limneticus* during the summer and *Melosira granulata* in the spring. Many species identified in phytoplankton samples were probably benthic or periphyton forms that had become resuspended into the water column. Ubiquitous but nonabundant taxa include the diatoms *Achnanthes minutissimia*, *Navicula radiosa*, *Rhioicosphenia curvata*, and *Surirella ovata*. The most common diatoms, however, were true planktonic forms.

Effects of river structure on the phytoplankton community

The abundance and composition of the phytoplankton community differed according to their locations along the Ohio River (Fig. 4). The annual averages of total cell numbers in the Ohio differed along the river course, with the McAlpine pool having the lowest overall densities (pairwise t test: $P < 0.05$). Cannelton pool near Louisville had consistently more resuspended benthic forms in the water column than the upper two regions. Patterns in densities also differed according to taxonomic group. Cyanobacterial numbers reflected the general trend of total cell density, but densities of greens and chrysophytes tended to increase further downstream. Considered during four separate temperature–flow periods (see Methods), certain patterns emerge (Table 3). The abundances of phytoplankton within most ecological and taxonomic groups changed significantly on progressing downstream and during most periods of the annual cycle. Perhaps the most

© 1997 NRC Canada

Fig. 3. Densities of the 10 most common algal and cyanobacterial species observed in the Ohio River during four seasons during 1991–1992. Bars represent means + 1 SE (n = 72 for each).

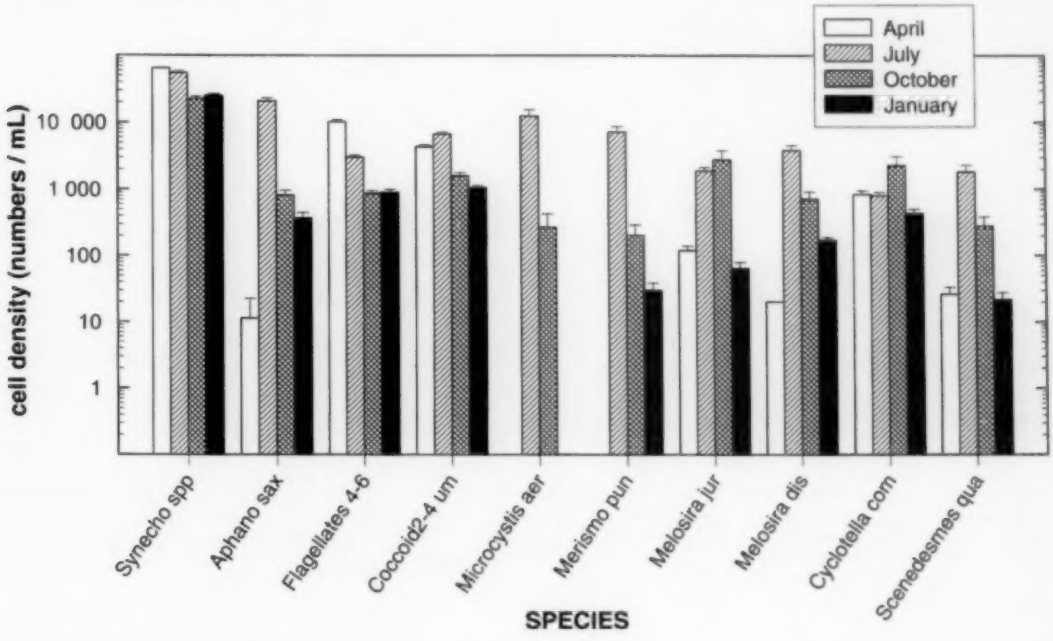
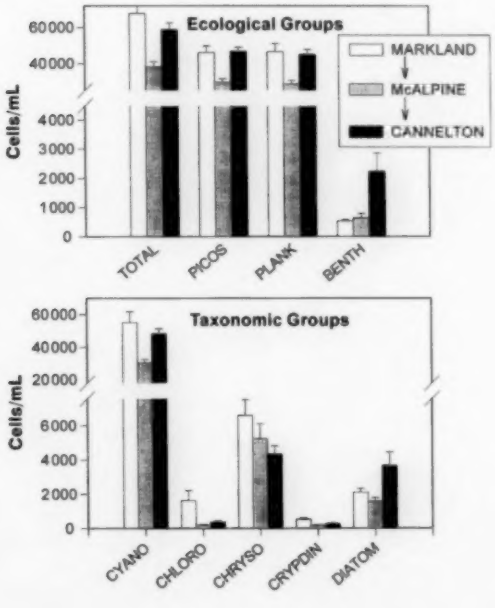


Fig. 4. Differences in the densities of phytoplankton in the Ohio River according to regional river pool. Data are classified into four ecological and five taxonomic groups (see Methods). Bars represent means + 1 SE (n = 72 for each).



pronounced difference was that during each period examined, total densities in the Ohio were consistently greatest in the upper Markland region and consistently lowest in midriver samples within the McAlpine pool (also lowest variation among sites).

Specific patterns suggest that during high-flow conditions in January, phytoplankton communities were most similar among river regions. In July, interpool differences were most pronounced. Markland pool had the highest numbers of total cells ($P < 0.001$), but the lowest numbers of resuspended benthic forms and diatoms ($P < 0.05$ and $P < 0.01$, respectively). Abundances of a few planktonic taxa were significantly lower in Markland, including *Melosira distans* and several *Navicula* species. In none of the general ecological or taxonomic categories were densities greatest in the downstream Cannelton pool. However, a bloom of *Microcystis aeruginosa* and *Melosira jurgensii* in Cannelton exceeded densities observed elsewhere in the Ohio ($P < 0.01$ for each).

Although navigation dams exerted some significant effects on water chemistry conditions in the Ohio, they had fewer significant effects on phytoplankton community structure (Fig. 5). The greatest general effect was a threefold decrease in the abundances of resuspended benthic forms in pools near the dams (ANOVA: $P < 0.001$). This pattern was also reflected in a reduction in the average diatom density at these sites. The effects of navigation dams were most pronounced during the summer, when temperatures were greatest and flow conditions were at a minimum (Table 4). July was the only month during which total cell densities (and most other categories) declined in the low pools near the navigation dams. In a few instances,

© 1997 NRC Canada

Table 3. Contrasts among three regions of the Ohio River (pools: Markland, McAlpine, and Cannelton) on phytoplankton abundance (see Methods for details).

	April		July		October		January	
	F	*P*	*F*	*P*	*F*	*P*	*F*	*P*
Community								
Total cells	12.74	<0.001	15.84	<0.001	17.26	<0.001	7.24	0.001
Total >2 μm	35.44	<0.001	25.35	<0.001	18.17	<0.001	3.06	ns
Planktonic species	17.13	<0.001	10.96	<0.001	16.36	<0.001	8.98	<0.001
Planktonics >2 μm	2.24	ns	14.32	<0.001	12.44	<0.001	7.17	0.001
Benthic species	48.24	<0.001	11.02	<0.001	19.21	<0.001	18.34	<0.001
Picoplankton	17.54	<0.001	9.87	<0.001	17.66	<0.001	10.05	<0.001
Taxonomic								
Cyanobacteria	17.66	<0.001	16.07	<0.001	18.96	<0.001	8.84	<0.001
Cyanobacteria >2 μm	3.15	0.049	22.56	<0.001	19.28	<0.001	0.72	ns
Chlorophyta	3.10	ns	30.29	<0.001	10.60	<0.001	2.13	ns
Chrysophyta	30.95	<0.001	5.17	0.008	None		12.46	<0.001
Crypto–Dinophyta	7.53	0.002	14.05	<0.001	24.38	<0.001	10.48	<0.001
Bacillariophyta	8.02	0.001	13.71	<0.001	16.80	<0.001	15.53	<0.001

Note: Phytoplankton responses are grouped into major categories on the basis of ecological and taxonomic properties (defined in Methods); differences are based on ANOVA; none, no cells of this category observed; *F*, calculated *F* ratio; *P*, probability; ns, nonsignificant, $P > 0.05$; data were log transformed to correct for normality.

Fig. 5. Effects of navigation dams on densities of phytoplankton in the Ohio River. Upper pools are main river sites (BTR) above the influence of the dams; lower pools are main river sites (LOW) at the dams (see Fig. 4 for additional details).

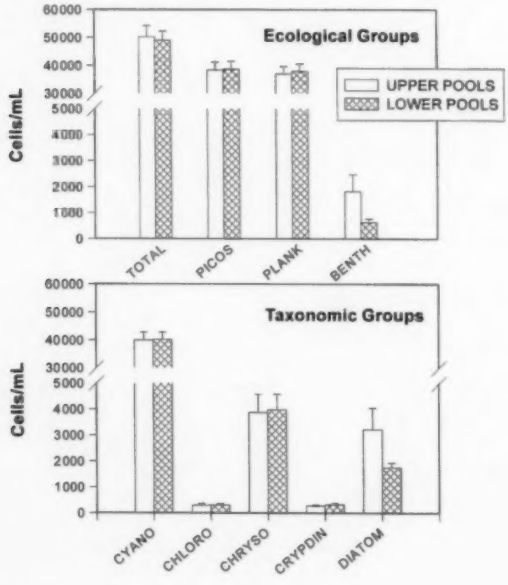
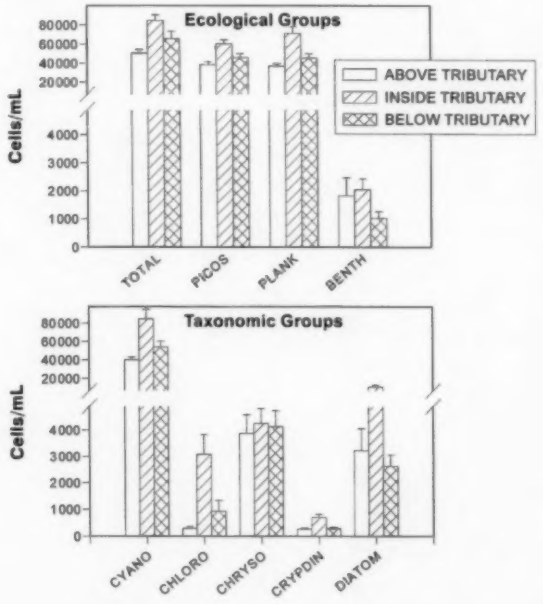


Fig. 6. Effects of major tributary rivers on densities of phytoplankton in the Ohio River. Sites are main river locations above, within, and downstream of tributaries at each regional pool (see Fig. 4 for additional details).



cell abundances significantly increased in these sites. During January, the density of larger cells and colonies (all those >2 μm) and the density of chrysophytes increased near navigation dams.

In nearly all cases examined, tributaries (ITR) had significantly greater densities of most phytoplankton species and groups than were observed in the main river (Fig. 6). This was especially true of diatoms and green algae; on average, tributaries contained 10-fold and 15-fold greater numbers of cells

© 1997 NRC Canada

Table 4. Comparisons of phytoplankton communities from above (designated main river below tributary, BTR) and below low-head navigation dams (designated main river, LOW) at each of three dam locations on the Ohio River.

	April		July		October		January	
	t	*P*	*t*	*P*	*t*	*P*	*t*	*P*
Community								
Total cells	–0.46	ns	–3.47	0.001	0.50	ns	1.55	ns
Total >2 µm	0.44	ns	–3.45	0.002	0.41	ns	2.13	0.040
Planktonic species	–0.33	ns	–2.73	0.010	0.70	ns	1.58	ns
Planktonics >2 µm	0.44	ns	–1.14	ns	1.36	ns	0.39	ns
Benthic species	–3.17	0.003	–3.32	0.002	–3.36	0.002	–3.96	<0.001
Picoplankton	–0.39	ns	–2.88	0.007	0.33	ns	1.55	ns
Taxonomic								
Cyanobacteria	–0.39	ns	–3.30	0.002	0.07	ns	1.60	ns
Cyanobacteria >2 µm	0.37	ns	–3.42	0.002	0.90	ns	1.30	ns
Chlorophyta	0.08	ns	–1.65	ns	0.83	ns	–2.90	0.012
Chrysophyta	0.51	ns	–3.16	0.003	None		2.02	0.051
Crypto/Dinophyta	–0.50	ns	0.66	ns	1.47	ns	0.20	ns
Bacillariophyta	0.52	ns	–1.80	ns	0.87	ns	–0.22	ns

Note: Phytoplankton responses are grouped into major categories on the basis of ecological and taxonomic properties (defined in Methods); differences are based on independent Student *t* tests with pooled variances; none, no cells of this category observed; *t*, calculated *t* value comparing upstream (BTR) versus at the dam (LOW); a positive *t* score indicates that densities increased at the dam sites of the main river; a negative score indicates a significant reduction; *P*, probability; ns, nonsignificant, $P > 0.05$.

Table 5. Comparisons of phytoplankton communities from above (designated ATR) and below (designated BTR) three major tributaries entering three regions of the Ohio river.

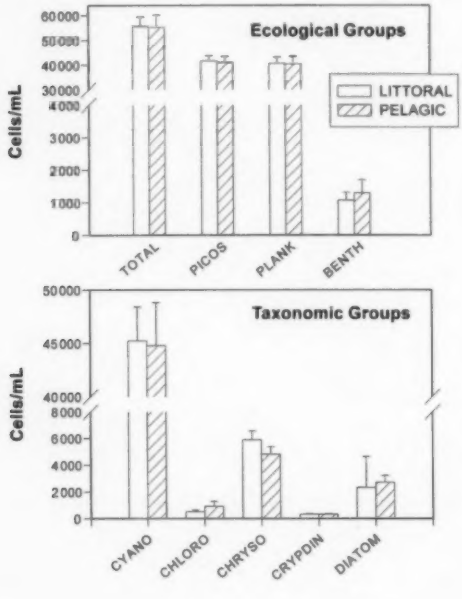
	April		July		October		January	
	t	*P*	*t*	*P*	*t*	*P*	*t*	*P*
Community								
Total cells	–0.55	ns	2.46	0.019	1.34	ns	–0.68	ns
Total >2 µm	1.52	ns	2.01	0.043	2.18	0.036	0.14	ns
Planktonic species	0.34	ns	2.27	0.029	1.26	ns	–0.84	ns
Planktonics >2 µm	0.91	ns	1.29	ns	2.02	ns	0.24	ns
Benthic species	–0.42	ns	–0.63	ns	0.34	ns	0.80	ns
Picoplankton	0.32	ns	2.25	0.031	1.20	ns	–0.85	ns
Taxonomic								
Cyanobacteria	0.33	ns	2.74	0.010	1.22	ns	–0.73	ns
Cyanobacteria >2 µm	0.48	ns	3.21	0.003	0.25	ns	2.46	0.020
Chlorophyta	0.97	ns	1.54	ns	0.98	ns	–1.57	ns
Chrysophyta	1.80	ns	1.55	ns	None		–0.79	ns
Crypto/Dinophyta	–1.43	ns	0.16	ns	1.22	ns	–0.49	ns
Bacillariophyta	0.08	ns	0.39	ns	1.65	ns	–0.15	ns

Note: Phytoplankton responses are grouped into major categories on the basis of ecological and taxonomic properties (defined in Methods); differences are based on independent Student *t* tests with pooled variances; none, no cells of this category observed; *t*, calculated *t* value comparing above versus below sites; a positive *t* score indicates that densities increased in the main river below the tributaries; a negative score indicates a significant reduction; *P*, probability; ns, nonsignificant, $P > 0.05$.

than the main channel of the Ohio. The annual averages indicate, however, only small effects on downstream (BTR) sites. For example, despite consistently large differences in absolute densities of diatoms in ATR versus ITR sites, sites downstream of tributaries (BTR) experienced no significant increases during any of the four seasons considered (Table 5). Green algae, chrysophytes, and cryptomonads–dinoflagellates were also unaffected by inputs from tributaries. This was true

© 1997 NRC Canada

Fig. 7. Differences in the densities of phytoplankton in the Ohio River in nearshore littoral habitats and open river pelagic habitats (see Methods for details of sampling locations; see Fig. 4 for additional details).



also of total cell densities in three of four seasons as well. Of the few significant effects of tributaries on downstream phytoplankton communities, most occurred during the summer. Planktonic forms, picoplankton, and cyanobacteria increased significantly below tributaries only during July. Most of this change was associated with increases in densities of the picoplankter *Synechococcus* sp. (t test: $P = 0.031$) and two large colonial forms, *Aphanothece saxicola* (t test: $P = 0.002$) and *Chroococcus limneticus* (t test: $P = 0.030$). Among 30 species of diatoms observed during the summer period, densities of only the small centric *Cyclotella meneghianna* were greater below tributaries (t test: $P = 0.026$). In January, only the larger celled and colonial cyanobacteria increased in sites downstream of tributaries.

Finally, we collected samples from midriver (pelagic) and nearshore (littoral) habitats to determine if the shallower, slower flowing conditions were perhaps more amenable to plankton development in the river. When viewed over the entire year, there were no significant differences in the densities of any ecological or taxonomic group between these two habitats (Fig. 7). The averages for pelagic and littoral densities within each category varied within ±20% of each other riverwide. With one exception, subsequent analyses of differences (via t tests) also indicated no differences among these two habitats ($P > 0.05$) for all individual seasons. During April, chrysophyte densities were significantly greater in littoral habitats than in the midriver pelagic zone. This difference was associated solely with 2.4-fold greater numbers of an unidentified tiny golden flagellate (4 μm wide × 6 μm long) in littoral habitats (t test: $P < 0.034$). Densities of all other taxa were insignificantly different between these two habitats.

Effects of physicochemical factors on Ohio River phytoplankton

Many of the temporal and spatial patterns observed for the phytoplankton community may be in part the result of changes in physical and chemical variables described earlier (Tables 1 and 2). To consider whether specific factors may be responsible for regulating densities in the Ohio River, a two-step analysis was used. First, correlations between chemical and biological variables were used to identify the primary factors probably influencing numbers of phytoplankton in certain ecological and taxonomic categories. Second, multiple regressions were used to determine the best quantitative model for predicting phytoplankton numbers.

Variations in many of the major physical and chemical variables measured correlated significantly with differences in densities of several phytoplankton groups in the Ohio River (Table 6). The most consistent pattern we observed with pairs of variables was with temperature; cell densities in all 12 phytoplankton groups correlated positively with greater temperatures (all $P < 0.001$). Among the algal nutrients, DIP concentrations correlated negatively with 10 phytoplankton variables, and inorganic N:P ratios correlated positively with nine phytoplankton variables. Larger colonial cyanobacteria were not significantly correlated with either total DIN or N:P ratio. Certain ecological and taxonomic groups exhibited different patterns in their responses to the suite of variables considered. Perhaps owing to the very large proportion of cells <2 μm, total cells, total cyanobacteria, and picoplankton densities responded similarly, with strong positive temperature, N:P, and turbidity correlations and negative correlations with DIP. Other groups were more distinct. In particular, benthic forms correlated negatively with current velocity, but with none of the common nutrient measures. Chrysophytes also were somewhat unlike other groups, having a positive correlation with current velocity, turbidity, and DIN but not with NO_3-N. Even though some of the chrysophyte taxa possess Si scales, neither members of this group nor diatoms correlated with dissolved Si. Variations in diatom densities correlated with a few variables only weakly; the strongest were significant positive correlations with temperature and dissolved Mg.

These relationships were considered for three major groups using MLR. The most important predictor of total phytoplankton density in the Ohio was found by MLR to be temperature (Table 7). Greater water temperatures were associated with significantly greater total cell density. The relationship explained only about 15% of the total variation (Fig. 8). The complete MLR model predicted that greater DIP and SO_4-S concentrations and greater current velocities would have negative impacts on total cells. On the basis of microscopic examinations, most of the river turbidity was not caused by phytoplankton cells, but MLR nonetheless predicted greater numbers during periods of higher turbidity. The six-variable model explained about 60% of the total variation in total cell densities.

Densities of planktonic forms were apparently affected by factors similar to those affecting total cells: turbidity was the best single predictor, which explained about 20% of the variation in planktonic densities (Table 7; Fig. 8). The complete predictive model contained the same independent variables (predictors) as for total cells, but their order of importance differed slightly.

© 1997 NRC Canada

Table 6. Correlations between 12 physical and chemical variables and phytoplankton abundances represented in 12 taxonomic and ecological categories (see Methods) measured for the Ohio River.

Ecological categories

	Total cells		Total >2 µm		Planktonic		Planktonic>2 µm		Benthic		Picoplankton	
	r	*P*	*r*	*P*	*r*	*P*	*r*	*P*	*r*	*P*	*r*	*P*
Temperature	0.386	***	0.509	***	0.282	***	0.363	***	0.295	***	0.247	***
Current	ns		ns		ns		ns		–0.192	**	ns	
Turbidity	0.329	***	ns		0.443	***	ns		ns		0.497	***
DIP	–0.377	***	–0.368	***	–0.395	***	–0.425	***	ns		–0.362	***
NH_4-N	0.258	***	0.127	*	0.286	***	ns		ns		0.339	***
NO_3-N	–0.253	***	–0.207	***	–0.241	***	–0.194	**	ns		–0.226	***
DIN	ns		ns		ns		ns		ns		ns	
N:P ratio	0.311	***	0.302	***	0.311	***	0.334	***	ns		0.294	***
Si	ns		ns		ns		–0.133	*	ns		ns	
SO_4-S	–0.129	*	ns		–0.175	**	ns		ns		–0.198	**
Ca	0.188	**	0.185	**	0.195	**	0.206	***	ns		0.164	**
Mg	0.264	***	0.275	***	0.257	***	0.199	**	0.173	**	0.241	***

Taxonomic categories

	Cyano		Cyano >2 µm		Chloro		Chryso		Crypto–Dino		Bacillario	
	r	*P*	*r*	*P*	*r*	*P*	*r*	*P*	*r*	*P*	*r*	*P*
Temperature	0.361	***	0.408	***	0.519	***	0.520	***	0.499	***	0.286	***
Current	ns		–0.390	***	–0.342	***	0.155	*	–0.381	***	–0.122	*
Turbidity	0.339	***	–0.476	***	–0.249	***	0.303	***	–0.333	***	ns	
DIP	–0.408	***	–0.122	*	–0.274	***	–0.236	***	ns		–0.182	**
NH_4-N	0.262	***	–0.258	***	–0.132	*	0.516	***	ns		ns	
NO_3-N	–0.294	***	–0.136	*	ns		ns		0.148	*	ns	
DIN	ns		ns		ns		0.201	**	0.159	*	ns	
N:P ratio	0.292	***	ns		0.240	***	0.339	***	ns		0.222	***
Si	ns		–0.292	***	–0.222	**	ns		–0.204	*	ns	
SO_4-S	–0.156	**	ns		0.141	*	0.149	*	0.465	***	ns	
Ca	0.179	**	ns		0.261	***	0.215	**	0.129	*	0.251	***
Mg	0.226	***	ns		0.156	*	0.572	***	0.317	***	0.291	***

Note: *r*, Spearman's product-moment correlation coefficient; *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$.

Densities of benthic taxa suspended in the water column could not be precisely estimated. Temperature was again the most important factor affecting cell densities, but only described about 9% of the total variation in their numbers (Fig. 8). The MLR approach was also less effective at producing a model that reliably estimates densities of benthic taxa in the water column ($P < 0.001$, but $r^2 = 0.18$). A further difference in these results was that greater levels of DIN were predicted to cause a reduction in cell densities and DIP concentrations were unimportant. Current velocity was not selected in this model.

Discussion

Our data show that the Ohio River supports a diverse and substantial phytoplankton community throughout its length. Phytoplankton densities $\geq 10^4$ cells/mL are comparable with many other large rivers in North America (Søballe and Kimmel 1987), including the Arkansas (Williams 1964), Hudson (Marshall 1988), Mississippi (Lange and Rada 1993), and Ohio (Peterson and Stevenson 1989), and many European rivers such as the Rhine (Admiraal et al. 1994), Meuse (Descy et al. 1987), Severn (Swale 1969), Thames (Lack 1971), and Wye (Jones 1984). Surface (1–5 m) phytoplankton densities in the Rio Tapajós, a large clearwater river in the Amazon basin, are also within the range observed in the Ohio; densities are perhaps an order of magnitude less in blackwater sections of the Amazon proper (Sioli 1975). Typical cell concentrations observed in large rivers are comparable with moderately productive lentic ecosystems with well-established plankton-based food webs. The numerical predominance of picoplankton-sized cells in the Ohio is typical of many freshwater systems (Stockner 1988), although there are few reports from rivers. Marshall (1988) found that autotrophic picoplankton were also numerically dominant in the Hudson River, but constitute <5% of the total biovolume. Our estimates suggest that even during maximum picoplankton numbers, picoplankton

© 1997 NRC Canada

Table 7. Summary of MLR analyses used to determine the major factors affecting key phytoplankton densities in the Ohio River.

MLR analyses

	Total cells/mL (r^2 = 0.610)				Planktonic cells/mL (r^2 = 0.624)				Benthic cells/mL (r^2 = 0.18)			
Step	Independent variable	Slope	*t*	*P*	Independent variable	Slope	*t*	*P*	Independent variable	Slope	*t*	*P*
1	Temperature	0.96 ± 0.06	15.8	***	Turbidity	0.61 ± 0.04	14.4	***	Temperature	1.09 ± 0.15	7.23	***
2	DIP	–0.24 ± 0.06	–3.6	***	Temperature	0.79 ± 0.05	14.7	***	NH_4^+ -N	–0.58 ± 0.11	–5.29	***
3	Turbidity	0.59 ± 0.05	12.5	***	Current	–0.13 ± 0.02	–5.07	***	Turbidity	0.29 ± 0.10	2.79	**
4	Current	–0.14 ± 0.03	–5.1	***	Ca^{2+}	0.24 ± 0.05	5.11	***	DIN	–0.58 ± 0.19	3.04	**
5	SO_4-S	–0.26 ± 0.05	–4.7	***	SO_4-S	–0.21 ± 0.05	–4.46	***	SO_4-S	–0.28 ± 0.13	–2.18	*
6	Ca^{2+}	0.26 ± 0.05	–4.9	***	DIP	–0.19 ± 0.06	–3.23	**	*y* intercept	2.54 ± 1.20		
	y intercept	7.61 ± 0.45			*y* intercept	7.48 ± 0.41						

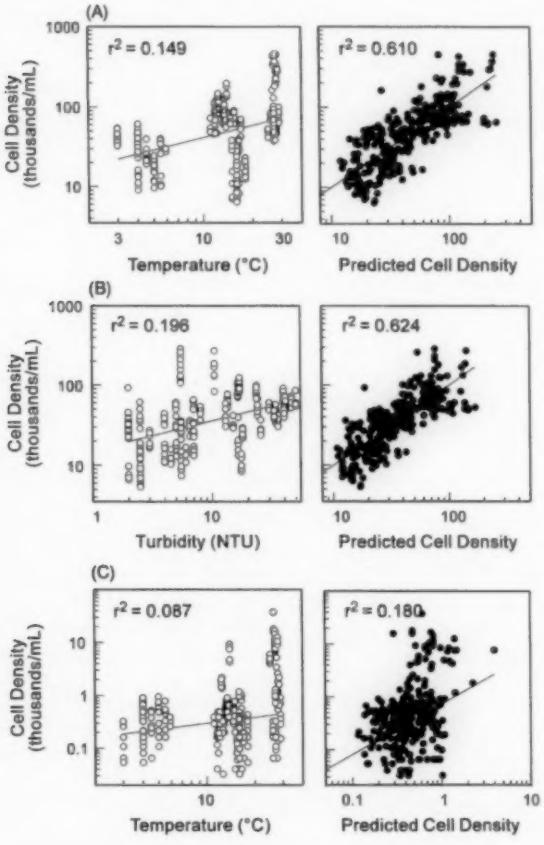
ANOVA

	Total cells/mL					Planktonic cells/mL					Benthic cells/mL				
Source	SS	df	MS	*F* ratio	*P*	SS	df	MS	*F* ratio	*P*	SS	df	MS	*F* ratio	*P*
Model	147.8	6	24.63	76.79	***	123.9	6	20.64	81.26	***	94.25	5	18.85	12.06	***
Residual	94.3	294	0.32			74.7	294	0.25			439.0	281	1.56		

Note: The independent variables tested were the 12 physical and chemical variables listed in Table 6. Significance of individual variables tested via Student's *t* test; r^2, coefficient of determination of the complete model; *P*, probability of a significant effect; *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$.

© 1997 NRC Canada

Fig. 8. Bivariate and multiple regressions predicting the density of (A) total, (B) planktonic, and (C) benthic cells in the Ohio River during 1991–1992. Independent variables in each bivariate regression are the factors selected by MLR as the primary factor affecting cell densities in each category; predicted density is the multivariable model established by MLR (see Table 7 for details).



cells represent only about 10% of the total summer phytoplankton biovolume in the Ohio.

A misunderstanding in aquatic ecology is the idea that large rivers lack a substantial phytoplankton community (Wetzel 1975; Junk et al. 1989). The view that self-sustaining algal populations are unimportant in large-order rivers may have been derived from the idea that most large rivers are deep and turbid; thus, phytoplankton production would be light limited most of the year (e.g., Vannote et al. 1980). Early reports suggested that much of the presumed phytoplankton community was either resuspended benthic species or cells washed in from pools or tributaries connected to lakes. Our data indicate that few (≈10% by species, 5% by numbers) of the Ohio River taxa were resuspended benthic forms (such as biraphid pennate diatoms) or tychoplanktonic species from littoral areas (e.g., *Mougeotia* sp.). The literature also suggests that lower reaches of rivers may have considerable in situ primary production, with associated planktonic and microbial food webs (Sedell et al. 1989; Cole et al. 1991; DeRuyter VanSteveninck et al. 1992). This production is supported in many large rivers by floodplain and anthropogenic inputs of nutrients (Friedrich and Müller 1984; Junk et al. 1989). Lack (1971) regarded the River Thames essentially as a series of nutrient-enriched impoundments with short retention times. Regions of the upper Mississippi where navigation dams have been constructed also support considerable phytoplankton communities, similar to levels observed in stratified lakes (Lange and Rada 1993). A detailed production budget for the River Lot (France) indicates that despite considerable abiotic (mineral particles) and allochthonous (detrital) light attenuation, navigation dams and water flow regulation favor the development of large phytoplankton populations during many months of the year (Decamps et al. 1984). We contend that these features are typical of many large rivers worldwide.

We found the species composition of the Ohio River phytoplankton to consist predominantly of diatoms, representing more than 60% of the species total. This general result is commonly reported in many large rivers (e.g., Williams 1964; Peterson and Stevenson 1989; Admiraal et al. 1994) but may obscure important temporal patterns in the community. Periods of maximum phytoplankton concentrations in the Ohio, as with most large rivers, occur during the summer, when temperatures are warmer and discharge is reduced. Midsummer assemblages in the Ohio were dominated in numbers (and biovolume) by colonial cyanobacteria, especially *Aphanothece saxicola*, *Merismopedia punctata*, and *Microcystis aeruginosa*, along with unicellular *Synechococcus* spp. Chlorophytes (*Scendesmus* spp. and other members of the chlorococcales) also peaked at this time. A few diatom species (*Melosira* spp.) were also abundant in the summer, but at the summer phytoplankton peak, they contributed on average only about 5.6% by number and 28% by volume. Diatoms were numerically more important in the community during the late autumn and early spring, when densities of cyanobacteria averaged between half and a third of their midsummer levels.

These results parallel findings from some large rivers, such as the Wye (Jones 1984), Severn (Swale 1969), Mississippi (Baker and Baker 1981; Huff 1986), and Hudson (Marshall 1988). The nearby Mississippi, which later joins the Ohio, is of particular interest to our work. During most years, the river typically experiences a distinct low-flow period in the summer, which appears to support large blooms of cyanobacteria (especially *Aphanizomenon flos-aquae*). During other periods the river is dominated by centric diatoms (e.g., *Melosira granulata*, *Melosira italica*, and *Cyclotella meneghiniana*). However, during 1986, the summer phytoplankton community of the upper Mississippi (≈km 1110) continued to be dominated numerically and in biovolume by the diatoms *Melosira granulata*, *Stephanodiscus astrea*, and *Cyclotella* spp. (Lange and Rada 1993). The authors suggested that a higher summer discharge in 1986 may have enabled diatoms to remain suspended in the water column and reduced stratification, which favors gas-vacuolate cyanobacteria. In our study on the Ohio, *Aphanothece* and *Microcystis* were the predominant bloom-forming species during the summer of 1991. Earlier data (1960–1964) from the Ohio River near Louisville indicated a distinct bimodality in phytoplankton communities, with greens (chlorococcales dominant 26% of all dates) and cyanobacterial forms (*Microcystis* and other cyanobacteria 5% of all dates) dominant in the

© 1997 NRC Canada

summer (Nall 1965; Riley 1969). Diatom maxima were observed during the spring and late fall (*Melosira* spp. 46% of dates; *Cyclotella* + *Stephanodiscus* 9% of dates). The River Rhine also is dominated by centric diatoms, such as *Stephanodiscus* in spring, but this dominance continues through the summer with multiple peaks of more than one species from this genus (Admiraal et al. 1994). All other reports from the Ohio suggest that *Stephanodiscus* is common, but we did not find it abundant. One possible reason for its low numbers may be that larger sized diatoms (with rapid sinking rates) may require extended periods of low discharge to maintain populations within the euphotic zone. More recently (1979–1981), abundances of several cyanobacterial (*Anacystis* and *Oscillatoria*) and green algal (*Scendesmus*) genera were shown to be strongly correlated with higher temperatures (Peterson and Stevenson 1989). The abundances of other taxa (e.g., *Chlamydomonas* and *Navicula* spp.) were inversely related to temperature during those years.

Differences in the community among river pools

The three large river segments we studied were compared to examine longitudinal differences in phytoplankton communities on a broader landscape level. Assemblages differed significantly among these pools during all times of the year. One of the most consistent results was that abundances of most planktonic forms were greatest in the upper (Markland) region of the Ohio, while densities of resuspended benthic forms were consistently greatest (but still <10% of the total) in the lowest study region (Cannelton). At first consideration, this pattern may seem counterintuitive. The RCC (Vannote et al. 1980) and riverine productivity model (Thorp and Delong 1994) each predict a greater (if limited) importance of phytoplankton to food webs in lower (larger order) reaches of rivers. However, average discharge increased along this stretch of the Ohio by only 19% (Ma = 2726 m^3/s, Mc = 3032 m^3/s, and Ca = 3240 m^3/s), and most of it flows through a geologically constricted valley; river order does not change. Despite its 360-km length, the present study was not sufficient to evaluate continuum effects on plankton abundances. There are also important physical changes in the Cannelton reach, with the "falls of the Ohio" creating increased turbulence, while the lower portion of the Cannelton pool flows though a broader floodplain. Densities of chrysophytes (mainly flagellates 4–6 μm) and green algae (mainly *Scenedesmus*, *Chlorella*, and *Actinastrum*) declined in this section. Overall, however, the lowest cell densities of most phytoplankton groups, as well as total densities (about 30% less on average), were observed in the middle McAlpine pool. We observed no significant differences in major algal nutrients or physical factors within this section of the Ohio (Tables 1 and 2). However, during this same period, total zooplankton densities were significantly greater in the McAlpine pool than in both upriver and downriver sections, primarily the result of much greater numbers of copepod nauplii (Thorp et al. 1994). The largest decline (40%) in phytoplankton numbers (36 500 to 21 800 cells/mL) between the Markland and McAlpine pools (from Ma LOW to Mc ATR) was observed in the picoplankton. Copepod nauplii feed primarily on algae, especially smaller forms (Williamson 1991); hence, grazing pressure seems a plausible cause for the dramatic declines in phytoplankton in the McAlpine pool.

Effects of navigation dams

A prominent feature of the Ohio, like many large rivers worldwide, is the presence of low-head navigation dams, which may influence the biomass and community structure of plankton communities. While large (high head) dams used for reservoir construction and power generation have profound effects on current velocities, residence time, and other conditions (Thornton et al. 1990), impacts of low-head navigation dams are more subtle. Average current velocity at the McAlpine dam was significantly reduced, but low sites in the other two pools were apparently unaffected. Certain nutrients were significantly different near navigation dams. DIP levels were at times significantly greater (January–July) while at other times significantly less (October), particularly in the McAlpine pool. With reduced current velocity, nutrient (especially P) spiraling lengths are expected to be shortened, and biotic processes (e.g., bacterial remineralization) may enhance availability (Newbold et al. 1981). However, our data for DIN and dissolved Si indicate no clear patterns with respect to navigation dams. In contrast, phytoplankton strongly influence levels of dissolved N, P, and Si in the River Rhine (Admiraal et al. 1994).

Navigation dams had significant, but fairly subtle effects on selected parts of the phytoplankton community (Fig. 5; Table 4). No general pattern for the effect of dams was observed on total phytoplankton densities in the river, although numbers of all cells >2 μm were significantly greater in low pools near navigation dams in January. In contrast, during July, abundances of many phytoplankton groups were significantly reduced at these sites. Only one group was consistently affected by this factor in the Ohio. Densities of benthic species were significantly lower just above low-head dams on all dates and among all sites. This suggests that small (and sometimes statistically nonsignificant) reductions in current velocity may allow bottom-dwelling species to settle out of the water column, even though this may have negligible effects on the total community. Also, sites just above navigation dams tend to be much deeper than other locations on the Ohio. This result is similar to results presented for the upper Mississippi and the River Wye in which heavy-walled diatoms are kept suspended in the water column during periods of high discharge (Jones 1984; Lange and Rada 1993). It also indicates that the current velocities in most parts of the Ohio usually are great enough to maintain a small assemblage of dislodged cells from solid substrates and river sediments. Thus, changes in discharge and navigation dams may have complex effects on river phytoplankton. Resuspension may increase total cell (especially benthic) densities, but can also dilute planktonic numbers. Descy et al. (1987) developed a model that predicts that seasonal variations in growth rates of river phytoplankton may affect their ability to compensate for the dilution effects of increased discharge. Further studies on the effects of navigation dams in large rivers must also take into account different growth rates of various phytoplankton species, which in general are known to vary inversely with cell size (Raven 1987).

Effects of tributaries

We also consider whether planktonic communities in the Ohio consist of resident populations or are washed in from lakes and rivers fed by three major tributaries (Miami,

© 1997 NRC Canada

Kentucky, and Salt rivers). If tributaries do not significantly alter the biomass or composition of river plankton, we may assume that the phytoplankton community is sustained by reproducing populations from the main channel and backwaters, rather than by renewal from other sources. The communities inside these tributaries differed significantly from the main river in several aspects. Tributary rivers tended to have greater total densities of phytoplankton (≈60% greater), with a greater proportion consisting of colonial cyanobacteria (50% greater), green algae (threefold greater), and diatoms (fivefold greater). Picoplankton-sized cells were less important in these smaller rivers. Despite large differences in densities of certain forms, effects of these tributaries on the main river were not large. Total densities of suspended cells in the Ohio above and below these tributaries were insignificantly different year-round. However, during the summer and fall, certain larger forms such as colonial cyanobacteria (e.g., *Aphanocapsa saxicola*) and some diatom species (e.g., *Cyclotella meneginiana*) were found in significantly greater densities downstream of tributaries. Tributaries also had few important effects on zooplankton communities in the Ohio (Thorp et al. 1994), with the exception of greater densities of rotifers (≈40% increase) and cladocerans (twofold increase). We suspect that the lack of strong tributary effects on Ohio plankton assemblages is the result of considerably larger total flow in the main river than in the smaller tributaries. For example, the average discharge of the Kentucky River (enters at McAlpine Pool) is ≈240 m^3/s (≈8% of total), compared with 3032 m^3/s in the Ohio. The Salt River (enters at Cannelton Pool) contributes only an additional ≈50 m^3/s. The communities in the main river would appear to consist of resident populations. In other large rivers, rapid doubling rates during the spring and summer (0.5–1.1/day) are able to sustain populations despite considerable downstream drift (Descy et al. 1987; DeRuyter VanSteveninck et al. 1992). Studies on the balance between dilution rates and growth rates of dominant species are necessary to further establish the importance of autotrophy in the lower sections of the Ohio.

Pelagic and littoral communities in the Ohio River

The flood-pulse concept introduced the idea that large rivers can maintain substantial in situ production in part by receiving nutrient and particulate inputs from flooded littoral and terrestrial (riparian) areas (collectively the aquatic–terrestrial transition zone, or ATTZ) adjacent to the river channel (Junk et al. 1989); these authors predicted that primary production in littoral and ATTZ areas should be greater than in the main channel. In contrast with zooplankton, whose average total densities were roughly 60% greater in littoral versus pelagic sites of the Ohio (Thorp et al. 1994), phytoplankton densities showed almost no difference with respect to these habitats. Because sampling was entirely restricted to suspended (≈planktonic) forms, probable differences in periphytic and benthic species were not seen. Another factor in this portion of the Ohio is that nearly all the river is located within a geologically constricted valley, with small floodplains above Louisville, Ky. Changes in flow conditions over the year brought about considerable dilution effects with probably little import of ATTZ-resident materials or populations in the Ohio. We suspect that the pattern in periphyton production in these two habitat types may be quite different.

Effects of physical and chemical factors

MLR analysis revealed that temperature and current velocity were important regulators of total density and the density of major phytoplankton groups in the Ohio. These are common factors affecting phytoplankton communities in other large rivers, such as the Mississippi (Lange and Rada 1993), Meuse (Descy et al. 1987), and Rhine (Admiraal et al. 1994). Water residence time (inversely related to discharge) was of critical importance in a cross-system analysis of rivers, impoundments, and lakes (Søballe and Kimmel 1987). For example, algal cell densities per unit P were least in rivers, an effect that was thought to be caused by the short residence time in flowing waters. The lack of any positive dependence of phytoplankton standing crop on PO_4-P concentrations in the Ohio River, while also seen in the upper Mississippi (Lange and Rada 1993), is still somewhat surprising. Concentrations of DIP in the Ohio remained fairly high throughout the year (40–50 μg P/L; Table 2) and exhibited no obvious seasonal patterns (Thorp et al. 1994). It is possible that primary production in the Ohio was simply not nutrient limited and was affected primarily by the dilution effects of river discharge. The effect of interannual variations in flow conditions is also important, as shown in the Mississippi by Lange and Rada (1993). They found that extended high-flow periods may cause shifts in community composition and can reduce interpool differences within the river. An obvious summer decline in dissolved Si in our study suggests biotic depletion, as has been observed by others (Admiraal et al. 1994; Descy et al. 1987; DeRuyter VanSteveninck et al. 1992), but not universally (Lange and Rada 1993). However, diatoms did not feature strongly in the Ohio phytoplankton community during 1991–1992, and both MLR and correlation analysis did not identify Si concentrations as a significant variable related to algal, or even diatom, densities. Other chemical factors, such as SO_4-S (– effect) and Ca^{2+} (+ effect), were also included in these empirical models, although their quantitative importance in explaining variation in phytoplankton densities was minor. We know of no studies that document the influence of SO_4-S on river phytoplankton, although Brasfield (1972), also using multiple regression, found that elevated SO_4-S in the Gallinas River (New Mexico) is strongly associated with greater levels of coliform bacteria. The Ca content of major rivers varies widely, and algal species requirements also differ markedly, but an earlier study of major phytoplankton genera in the Ohio (Peterson and Stevenson 1989) did find positive correlations between certain major taxa and greater total alkalinity, which typically corresponds to higher Ca^{2+} levels. Moss (1988) also commented that certain algal groups, such as desmids (rare in the Ohio River), are strong calciphobes.

While physical factors (temperature and flow) appear to have had the greatest impact on phytoplankton communities, our data suggest that biotic regulation by grazing zooplankton may also play an important role. Experimental studies are needed that can tease apart the influences of herbivory, nutrient supply, and flow regime on the growth rates of primary producers in large rivers. Further studies are also needed to examine the coupling between the resuspension of benthic and periphytic forms on the total biomass of cells suspended in the

© 1997 NRC Canada

water column. These connections may enable the river to maintain a positive autotrophic balance during winter and spring when discharge is high and temperatures are reduced.

Acknowledgments

We gratefully acknowledge the efforts and assistance of Wei Fang, Kim Greenwood, and Dr. Kim H. Haag in collecting plankton and water chemistry samples during this study. We also thank Lori Campbell for assistance with water chemistry analyses. J.H.T. was supported by a grant from the National Science Foundation (NSF) (DEB-9307225) and J.D.W. by the Routh Endowment Fund and the NSF (DIR-9002145).

References

Admiraal, W., Breebaart, L., Tubbing, G.M.J., VanZanten, B., DeRuitjer VanSteveninck, E.D., and Bijkerk, B. 1994. Seasonal variation in composition and production of planktonic communities in the lower River Rhine. Freshwater Biol. **32**: 519–531.

American Public Health Association. 1985. Standard methods for the analysis of water and wastewater. 16th ed. American Public Health Association, Washington, D.C.

Baker, A.L., and Baker, K.K. 1981. Seasonal succession of the phytoplankton in the upper Mississippi River. Hydrobiologia, **83**: 295–301.

Bran+Luebbe Analyzing Technologies. 1986*a*. Ortho phosphate in water and seawater. Industrial method No. 812-86T. Bran+Luebbe Inc., Buffalo Grove, Ill.

Bran+Luebbe Analyzing Technologies. 1986*b*. Ammonia in water and seawater. Industrial method No. 804-86T. Bran+Luebbe Inc., Buffalo Grove, Ill.

Bran+Luebbe Analyzing Technologies. 1987*a*. Nitrate/nitrite in water and seawater. Industrial method No. 818-87T. Bran+Luebbe Inc., Buffalo Grove, Ill.

Bran+Luebbe Analyzing Technologies. 1987*b*. Silicates in water and wastewater. Industrial method No. 785-86T. Bran+Luebbe Inc., Buffalo Grove, Ill.

Bran+Luebbe Analyzing Technologies. 1989. Sulfate in water and wastewater. Industrial method No. 847-87T. Bran+Luebbe Inc., Buffalo Grove, Ill.

Brasfield, H. 1972. Environmental factors correlated with size of bacterial populations in a small polluted stream. Appl. Microbiol. **24**: 349–352.

Cole, J.J., Caraco, N.F., and Peierls, B. 1991. Phytoplankton primary production in the tidal, freshwater Hudson River, New York (USA). Verh. Int. Ver. Theor. Angew. Limnol. **24**: 1715–1719.

Cushing, C.E. 1967. Periphyton productivity and radionuclide accumulation in the Columbia River, Washington, USA. Hydrobiologia, **29**: 125–139.

Decamps, H., Capblanq, J., and Tourenq, J.N. 1984. Lot. *In* Ecology of European rivers. *Edited by* B.A. Whitton. Blackwell, Oxford. pp. 207–235.

DeRuyter VanSteveninck, E.D., Admiraal, W., Breebaart, L., Tubbing, G.M.J., and van Zanten, B. 1992. Plankton in the River Rhine: structural and functional changes observed during downstream transport. J. Plankton Res. **14**: 1351–1368.

Descy, J.-P., Servais, P., Smitz, J.S., Billen, G., and Everbecq, E. 1987. Phytoplankton biomass and production in the River Meuse (Belgium). Water Res. **21**: 1557–1566.

Friedrich, G., and Müller, D. 1984. Rhine. *In* Ecology of European rivers. *Edited by* B.A. Whitton. Blackwell, Oxford. pp. 265–315.

Huff, D.R. 1986. Phytoplankton communities in navigation pool No. 7 of the upper Mississippi River. Hydrobiologia, **136**: 47–56.

Hynes, H.B.N. 1970. The ecology of running waters. University of Toronto Press, Toronto, Ont.

Hynes, H.B.N. 1989. Keynote address. *In* Proceedings of the International Large River Symposium (LARS). *Edited by* D.P. Dodge. Can. Spec. Publ. Fish. Aqaut. Sci. No. 106. pp. 5–10.

Jones, F.H. 1984. The dynamics of suspended algal populations in the lower Wye catchment. Water Res. **18**: 25–35.

Junk, W.J., Bayley, P.B., and Sparks, R.E. 1989. The flood-pulse concept in the river–floodplain systems. *In* Proceedings of the International Large River Symposium (LARS). *Edited by* D.P. Dodge. Can. Spec. Publ. Fish. Aquat. Sci. No. 106. pp. 110–127.

Kofoid, C.A. 1903. The plankton of the Illinois River, 1894–1899, with introductory notes upon the hydrography of the Illinois River and its basin. Part 1. Quantitative investigations and general results. Bull. Ill. State Lab. Nat. Hist. **6**: 95–629.

Lack, T.J. 1971. Quantitative studies on the phytoplankton of the Rivers Thames and Kennet at Reading. Freshwater Biol. **1**: 213–224.

Lange, T.R., and Rada, R.G. 1993. Community dynamics of phytoplankton in a typical navigation pool in the upper Mississippi River. J. Iowa Acad. Sci. **100**: 21–27.

Lowe, R.L. 1974. Environmental requirements and pollution tolerance of freshwater diatoms. U.S. E.P.A. Environ. Mon. Ser. No. EPA-670/4-74-005.

Luttenton, M.L., Vansteenburg, J.B., and Rada, R.G. 1986. Phycoperiphyton in selected reaches of the Upper Mississippi River: community composition, architecture, and productivity. Hydrobiologia, **136**: 31–46.

Marshall, H.G. 1988. Seasonal phytoplankton composition and concentration patterns within the Hudson River. Final report. Hudson River Foundation, New York.

Moss, B. 1988. Ecology of fresh waters. 2nd ed. Blackwell, Oxford.

Nall, R.W. 1965. Some factors affecting phytoplankton pulses in the Ohio River at Louisville, Kentucky. Ph.D. dissertation, University of Louisville, Louisville, Ky.

Newbold, J.D., Elwood, J.W., O'Neill, R.V., and VanWinkle, W. 1981. Measuring nutrient spiralling length in streams. Can. J. Fish. Aquat. Sci. **38**: 860–863.

Peterson, C.G., and Stevenson, R.J. 1989. Seasonality in river phytoplankton: multivariate analyses of data from the Ohio River and six Kentucky tributaries. Hydrobiologia, **183**: 99–114.

Raven, J.A. 1987. Physiological consequences of extremely small size for autotrophic organisms in the sea. *In* Photosynthetic picoplankton. *Edited by* T. Platt and W.K.W. Li. Can. Bull. Fish. Aquat. Sci. No. 214. pp. 1–70.

Riley, M.B. 1969. Some observations of the phytoplankton of the McAlpine pool of the Ohio River. M.S. dissertation, University of Louisville, Louisville, Ky.

Sedell, J.R., Richey, J.E., and Swanson, F.J. 1989. The river continuum concept: a basis for the expected ecosystem behavior of very large rivers? *In* Proceedings of the International Large River Symposium (LARS). *Edited by* D.P. Dodge. Can. Spec. Publ. Fish. Aquat. Sci. No. 106. pp. 49–55.

Sioli, H. 1975. Tropical river: the Amazon. *In* River ecology. *Edited by* B.A. Whitton. Blackwell, Oxford. pp. 461–488.

Søballe, D.M., and Kimmel, B.L. 1987. A large-scale comparison of factors influencing phytoplankton abundance in rivers, lakes, and impoundments. Ecology, **68**: 1943–1954.

Sokol, R.R., and Rohlf, F.J. 1995. Biometry. 3rd ed. W.H. Freeman & Co., New York.

Stockner, J.G. 1988. Phototrophic picoplankton: an overview from marine and freshwater ecosystems. Limnol. Oceanogr. **33**: 765–775.

Swale, E.M.F. 1969. Phytoplankton in two English rivers. J. Ecol. **57**: 1–23.

Thornton, K.W., Kimmel, B.L., and Payne, F.E. (*Editors*). 1990. Reservoir limnology: ecological perspectives. Wiley, New York.

© 1997 NRC Canada

Thorp, J.H., and Delong, M.D. 1994. The riverine productivity model: an heuristic view of carbon sources and organic processing in large river ecosystems. Oikos, **70**: 305–308.

Thorp, J.H., Black, A.R., Haag., K.H., and Wehr, J.D. 1994. Zooplankton assemblages in the Ohio River: seasonal, tributary, and navigation dam effects. Can. J. Fish. Aquat. Sci. **51**: 1634–1643.

Utermöhl, H. 1958. Zur Vervollkommnung der quantitativen Phytoplankton-Methodik. Mitt. Int. Ver. Theor. Angew. Limnol. **9**: 1–38.

Vannote, R.L., Minshall, G.W., Cummins, K.W., Sedell, J.R., and Cushing, C.E. 1980. The River Continuum Concept. Can. J. Fish. Aquat. Sci. **37**: 130–137.

Wetzel, R.G. 1975. Primary production. *In* River ecology. *Edited by* B.A. Whitton. Blackwell, Oxford. pp. 230–247.

Whitton, B.A. 1975. Algae. *In* River ecology. *Edited by* B.A. Whitton. Blackwell, Oxford. pp. 81–105.

Wilkinson, L. 1992. SYSTAT for Windows: statistics, version 5 edition. SYSTAT, Inc., Evanston, Ill.

Williams, L.G. 1964. Possible relationships between plankton–diatom species numbers and water quality estimates. Ecology, **45**: 809–823.

Williamson, C.L. 1991. Copepoda. *In* Ecology and classification of North American freshwater invertebrates. *Edited by* J.H. Thorp and A.P. Covitch. Academic Press, New York. pp. 787–822.

Zacharias, O. 1898. Das Potamoplankton. Zool. Anz. **21**: 41–48.

Appendix

Phytoplankton species identified from the Ohio River during 1991–1992 (pools and sites are arranged in downstream order from left to right (+, presence at site; cf., approximate identification); see Methods for explanation of site names and locations).

	Markland pool				McAlpine pool				Cannelton pool			
	ATR	ITR	BTR	LOW	ATR	ITR	BTR	LOW	ATR	ITR	BTR	LOW
Cyanobacteria												
Anabaena sp. A (3–4 μm)	+	+	+			+	+		+	+		
Aphanizomenon flos-aquae		+					+					
Aphanothece cf. *saxicola*	+	+	+	+	+	+	+	+	+	+	+	+
Chroococcus limneticus (cf.)	+	+	+			+				+	+	+
Chroococcus sp. (≈5 μm)		+	+					+	+	+		
Dactylococcopsis rhaphidioides	+	+	+	+	+	+	+	+	+	+	+	+
Lyngbya sp. A (1.9–2.5 μm)			+				+			+		+
Merismopedia punctata	+	+	+	+	+	+	+	+	+	+	+	+
Merismopedia tenuissima	+	+	+	+	+		+		+	+	+	+
Microcystis cf. *aeruginosa*	+	+	+	+	+	+	+	+	+	+	+	
Oscillatoria sp. (5 μm)	+			+								
Pseudanabaena catanata	+	+	+	+	+	+	+	+	+	+	+	+
Rhabdoderma cf. *minima*	+		+	+				+	+	+		
Synechococcus spp.	+	+	+	+	+	+	+	+	+	+	+	+
Coccoid cells (2–4 μm)	+	+	+	+	+	+	+	+	+	+	+	+
Chlorophyta												
Actinastrum cf. *gracilimum*	+	+	+	+	+	+	+		+	+	+	
Ankistrodesmus falcatus	+	+	+		+	+	+	+	+	+	+	+
Characium sp.		+	+									
Chlamydomonas sp. A (≈14 μm)	+	+	+	+	+	+	+	+	+	+	+	+
Chlorella sp. (≈13 μm)		+										
Coelastrum microporum (cf.)	+	+		+						+		
Dictyosphaerium ehrenbergianum		+	+								+	
Euastrum denticulatum (cf.)	+	+										+
Eudorina elegans			+	+					+			
Golenkinia radiata	+							+	+	+	+	+
Gonium pectorale	+	+	+	+	+		+	+	+	+	+	+
Haematococcus sp. (7.5 μm)		+								+		
Mougeotia sp.		+										
Oocystis sp.	+	+		+	+	+		+			+	+
Pandorina morum								+		+		
Pediastrum boryanum	+	+									+	+
Pediastrum duplex	+	+		+	+		+			+		
Scendesmus quadricauda	+	+	+	+	+	+	+	+	+	+	+	+
Scenedesmus dimorphus (cf.)	+	+	+	+	+	+	+			+	+	
Selenastrum cf. *westii*	+	+	+	+	+	+	+	+	+	+	+	+
Sorastrum spinulosum (cf.)	+	+		+						+		+
Staurastrum gracile (cf.)		+	+	+						+		
Staurastrum not *gracile*								+				
Euglenophyta												
Euglena cf. *tripteris*	+	+	+	+	+	+	+	+		+	+	+
Phacus sp. A							+					

© 1997 NRC Canada

Appendix (*continued*).

	Markland pool				McAlpine pool				Cannelton pool			
	ATR	ITR	BTR	LOW	ATR	ITR	BTR	LOW	ATR	ITR	BTR	LOW
Chrysophyta												
Dinobryon stipitatum (cf.)	+	+	+	+	+	+	+	+		+	+	+
Dinobryon divergens						+						
Mallomonas sp. (small)								+				
Ochromonas sp. (6 × 4 μm)										+	+	
Resting cyst type A						+				+		
Small flagellates (8–15 μm)	+	+	+	+	+	+	+	+	+	+	+	+
Synura sp. (11–15 μm/cell)						+				+		+
Tiny flagellates	+	+	+	+	+	+	+	+	+	+	+	+
Pyrrophyta–Cryptophyta												
Ceratium hirundinella		+	+	+								
Ceratium cysts		+	+	+	+	+		+		+		
Cryptomonas sp. A (25 μm)	+	+	+	+	+	+	+	+	+	+	+	+
Peridinium sp.	+	+	+	+	+	+	+	+	+	+	+	
Bacillariophyta												
Achnanthes lanceolata		+	+	+	+	+	+	+	+	+	+	+
Achnanthes minutissima	+	+	+	+	+	+	+	+	+	+	+	+
Amphipleura pellucida			+									
Amphora ovalis		+		+			+	+	+	+	+	+
Amphora perpusilla		+				+	+				+	
Asterionella formosa	+	+	+	+	+	+	+	+	+		+	+
Caloneis amphisbaena	+	+									+	
Caloneis venticosa (cf.)						+						
Cocconeis pediculus						+	+		+	+		+
Cocconeis placentula	+	+	+			+	+	+	+	+	+	+
Cyclotella comta	+	+	+	+	+	+	+	+	+	+	+	+
Cyclotella meneghiniana	+	+	+	+	+	+	+	+	+	+	+	+
Cyclotella ocellata (cf.)		+							+			+
Cymatopleura solea	+							+				
Cymbella amphicephala	+		+	+			+	+	+			+
Cymbella lunata			+			+		+		+	+	+
Cymbella minuta	+	+	+	+	+	+	+		+	+	+	+
Cymbella prostrata (cf.)	+							+				
Cymbella turgida			+	+							+	
Denticula tenuis (cf.)				+								
Diatoma anceps							+					
Diatoma vulgare	+	+	+	+	+	+	+		+	+	+	+
Diploneis elliptica (cf.)				+						+		
Eunotia exigua	+				+							
Eunotia pectinalis	+								+		+	
Eunotia perpusilla	+											+
Fragilaria crotonensis	+	+	+	+	+	+	+	+	+	+	+	+
Fragilaria virescens	+	+		+	+			+	+	+	+	+
Frustulia rhomboides	+			+	+	+						
Gomphoneis herculeana	+						+		+			
Gomphonema angustatum	+	+	+	+	+	+	+		+	+	+	+
Gomphonema subclavatum	+	+		+	+		+	+				
Gomphonema truncatum	+			+			+	+			+	+
Gyrosigma scalproides	+	+	+	+		+	+		+	+	+	+
Hantzschia amphioxys						+	+			+	+	
Melosira distans (small var.)	+	+	+	+	+	+	+	+	+	+	+	+
Melosira granulata	+	+	+	+	+	+	+	+	+	+	+	+
Melosira gran. v. *angustissima*	+	+	+	+	+	+	+	+	+	+	+	+
Melosira italica (cf.)	+	+							+			+
Melosira jurgensii (cf.)	+	+	+	+	+	+	+	+	+	+	+	+
Melosira varians	+	+	+	+	+	+	+	+	+		+	+
Meridion circulare	+	+	+	+	+				+	+	+	+
Navicula confervacea	+	+	+	+						+		+
Navicula decussis (cf.)	+	+	+	+	+	+	+			+	+	+
Navicula exigua (cf.)							+					

© 1997 NRC Canada

Appendix (*concluded*).

	Markland pool				McAlpine pool				Cannelton pool			
	ATR	ITR	BTR	LOW	ATR	ITR	BTR	LOW	ATR	ITR	BTR	LOW
Navicula halophila (cf.)		+		+	+						+	+
Navicula integra				+								
Navicula menisculus		+		+	+			+	+		+	
Navicula muticoides						+			+			
Navicula pupula v. *elliptica*	+		+	+	+							+
Navicula radiosa	+	+	+	+	+	+	+	+	+	+	+	+
Navicula reinhardtii (cf.)				+				+	+	+		+
Navicula sp. L			+									+
Navicula sp. Q			+									
Navicula sp. R	+	+	+	+	+	+	+	+	+	+	+	+
Neidium iridis v. *amphigomphus*	+											
Nitzschia acicularis (cf.)	+	+	+	+	+	+	+	+	+	+	+	+
Nitzschia actinastroides (cf.)	+	+				+				+		
Nitzschia dissipata	+		+			+			+	+		
Nitzschia dubia		+		+								
Nitzschia fonticola (cf.)	+	+	+	+	+	+	+	+	+	+	+	+
Nitzschia frustulum (cf.)	+	+	+	+	+	+	+	+	+	+		+
Nitzschia littoralis	+	+					+		+			+
Nitzschia palea (cf.)	+	+	+	+	+	+	+	+	+	+	+	+
Nitzschia sigma (cf.)	+		+	+	+		+		+		+	
Pinnularia borealis	+			+			+			+	+	
Pinnularia braunii (cf.)					+		+					
Pinnularia maior	+											
Pinnularia rupestris (cf.)	+											
Plagiotropis lepidoptera				+								
Rhoicosphenia curvata	+	+	+	+	+	+	+	+	+	+	+	+
Stauroneis anceps								+				
Stauroneis branderi				+							+	+
Stephanodiscus astraea	+	+	+	+	+	+	+	+	+	+	+	+
Surirella linearis		+	+	+			+	+	+	+	+	
Surirella ovata	+	+	+	+	+	+	+	+	+	+	+	+
Synedra delicatissima	+	+	+	+	+	+	+		+	+	+	
Synedra rumpens	+	+	+	+	+			+	+	+	+	+
Synedra ulna	+	+	+	+	+	+	+	+	+	+	+	+
Synedra ulna v. *amphirhynchus*	+									+	+	
Synedra ulna v. *contracta*	+		+	+			+	+				
Tabellaria fenestrata	+		+	+	+							

© 1997 NRC Canada

Genetic manipulation of sex ratio for the large-scale production of all-male tilapia, *Oreochromis niloticus*

G.C. Mair, J.S. Abucay, D.O.F. Skibinski, T.A. Abella, and J.A. Beardmore

Abstract: We describe the results of a breeding programme for Nile tilapia, *Oreochromis niloticus*, incorporating estrogen-induced sex reversal of male to female and progeny testing to generate novel 'YY' male genotypes. 'YY' male genotypes proved to be as viable and fertile as normal XY males and to sire a mean progeny sex ratio of 95.6% male. While these results confirm the hypothesis of predominantly monofactorial sex determination, there appears to be a genetic basis for the occurrence of occasional females in the progeny of 'YY' males. It is likely that these arise through the action of several autosomal sex modifying genes. We report the first instance of feminization of YY genotypes in tilapia and the mass production of 'YY' males through YY × YY matings, obviating the need for time-consuming progeny testing to discriminate XY and YY male genotypes. This enables the production of 'YY' males and their all-male progeny, known as genetically male tilapia, to be mass produced on a commercial scale. We conclude that the YY male technology provides a robust and reliable solution to the serious and widespread problem of early sexual maturation, unwanted reproduction, and overpopulation in tilapia culture.

Résumé : Nous décrivons les résultats d'un programme d'amélioration génétique du tilapia du Nil, *Oreochromis niloticus*, en incorporant l'inversion du sexe des mâles en femelles induite par les oestrogènes et l'analyse de la progéniture pour produire des nouveaux génotypes mâles 'YY'. Les mâles de génotype 'YY' se sont révélés aussi viables et fertiles que les mâles normaux XY et le sex-ratio moyen de leur progéniture comportait 95,6% de mâles. Bien que ces résultats confirmaient l'hypothèse d'une détermination du sexe à prédominance monofactorielle, il semble y avoir un fondement génétique pour l'apparition occasionnelle de femelles dans la progéniture des mâles 'YY'. Il est probable que ces femelles proviennent de l'action de plusieurs gènes autosomiques modifiant le sexe. Nous signalons le premier cas de féminisation de génotypes YY chez le tilapia et de production massive de mâles 'YY' par croisement YY × YY, ce qui rend inutile l'analyse de la progéniture, tâche longue et fastidieuse, pour distinguer les génotypes mâles XY et YY. Cette méthode permet la production massive à l'échelle commerciale de mâles 'YY' et de leur progéniture entièrement mâle, que l'on appelle tilapias génétiquement mâles. Nous avons conclu que la technologie mâle fournit une solution fiable et robuste au problème sérieux et répandu de la maturation sexuelle précoce, de la reproduction non voulue et de la surpopulation dans la culture du tilapia.
[Traduit par la Rédaction]

Introduction

The Nile tilapia, *Oreochromis niloticus*, is widely used in tropical aquaculture and is becoming an increasingly popular species for culture in environmentally controlled conditions in temperate countries. Aquaculture statistics for 1993 place annual world production for this species at > 365 000 metric tons (Food and Agriculture Organization of the United Nations 1995), which is likely to be an underestimate. Production is presently expanding rapidly, especially in Southeast Asia, and the Nile tilapia is now the principal species of choice in freshwater aquaculture in many tropical countries.

This species has been the focus of considerable biological research, with genetics receiving much emphasis (Pullin and Maclean 1992). Many studies have concentrated on the genetic basis of sex determination in *O. niloticus* and other *Oreochromis* species, as reviewed by Mair et al. (1991*a*, 1991*b*) and Trombka and Avtalion (1993). The interest in the genetics of sex determination in tilapia is motivated by the practical and commercial implications of the production of monosex progeny for use in aquaculture in addition to fundamental scientific curiosity.

Culture of monosex progeny, preferably males, which, in tilapia species, grow faster and to a larger size than females, has long been recognized as the most effective solution to the widespread problem of early sexual maturation and uncontrolled reproduction in tilapia culture. To date, this has been achieved either through manual sexing, direct hormonal sex reversal, or hybridization, the relative merits of which are reviewed in Mair and Little (1991) and Wohlfarth (1994). All of these techniques have significant disadvantages in their application, and none has become widely used in aquaculture, especially in developing countries. This is in contrast with the widespread culture of monosex female salmonids produced

Received January 9, 1996. Accepted July 16, 1996.
J13251

G.C. Mair,[1] D.O.F. Skibinski, and J.A. Beardmore. School of Biological Sciences, University of Wales Swansea, Swansea SA2 8PP, Wales, U.K.
J.S. Abucay and T.A. Abella. Freshwater Aquaculture Center, Central Luzon State University, Nueva Ecija 3120, Philippines.

[1] Author to whom correspondence should be sent at the following address: Freshwater Aquaculture Center, Central Luzon State University, Nueva Ecija 3120, Philippines. e-mail: GCMair@phil.gn.apc.org.

© 1997 NRC Canada

through genetic manipulation of sex determination (Bye and Lincoln 1986).

Although much studied, the precise mechanisms by which sex is determined in tilapia, specifically *O. niloticus*, are not fully understood. Early hypotheses were based on the sex ratios observed in hybrid crosses of different species, the most comprehensive of these hypotheses being that based on a theory of autosomal influence (Hammerman and Avtalion 1979). However, so far, no theory based on hybrid sex ratios successfully explains all observed sex ratios, which may be highly variable in some crosses such as those between *O. niloticus* and *Oreochromis aureus*. Recent research has concentrated on intraspecific sex ratios. Several authors have presented evidence that indicates that *O. niloticus* has a predominantly monofactorial mechanism of sex determination with heterogametic XY males and homogametic XX females (Mair et al. 1991*a*; Trombka and Avtalion 1993). However, this simple monofactorial hypothesis fails to explain some deviations from predicted sex ratios based on studies involving sex reversal and chromosome set manipulation. Hussain et al. (1994) hypothesized the existence of an autosomal sex-modifying locus (with alleles *SR* and *sr*) epistatic to the gonosomal locus and which induces female to male sex reversal when *sr* is homozygous. This hypothesis was developed to explain the occurrence of varying proportions of males in heterozygous and homozygous meiotic and mitotic gynogenetic progeny (Mair et al. 1991*a*; Hussain et al. 1994). However, this hypothesis still fails to explain some of the aberrant sex ratios observed in crosses of hormonally sex reversed fish, and it has been concluded that these arise through additional autosomal influences and (or) environmental influences (Mair et al. 1990; Trombka and Avtalion 1993). Baroiller et al. (1995) presented evidence for a temperature effect on sex differentiation. In putative all-female progeny from androgen sex-reversed males (XX) crossed with normal females, they observed significantly higher proportions of aberrant males in progeny reared at high temperatures (36°C) during the period of sex differentiation. Similar results have been obtained in our laboratory using different strains of *O. niloticus* (J.S. Abucay, unpublished data). The hypothesis of a temperature effect on sex determination still does not explain the occurrence of a small percentage of males in the putative monosex female progeny reared at ambient temperatures. Thus, currently available data indicate that sex determination in *O. niloticus*, while influenced by several factors, is best described as predominantly monofactorial, with an underlying mechanism of male heterogamety playing the major role.

On the basis of this hypothesis of predominantly monofactorial sex determination, several authors have proposed a breeding programme through which novel 'YY' males could be generated in *Oreochromis mossambicus* (Yang et al. 1980; Varadaraj and Pandian 1989) and in *O. niloticus* (Mair 1988; Baroiller and Jalabert 1989; Scott et al. 1989). These authors demonstrated the viability of very small numbers of 'YY' males, but none demonstrated that these could be produced and spawned on the large scale that would be necessary for the technology, termed the YY male technology, to be applied to aquacultural practice. Our objectives in this study were to further investigate sex determination in *O. niloticus* to determine the viability of, and the progeny sex ratios produced by, 'YY' males and to test the feasibility of large-scale production of all-male tilapia through genetic manipulation of sexual phenotype.

Materials and methods

Genetic origins of stocks and genetic nomenclature

The strain of *O. niloticus* used in these investigations, known in the Philippines as Egypt-Swansea, originates from Lake Manzala, Egypt, from where it was collected and transferred to the University of Stirling, Scotland, in 1979 (Hussain 1992). Several introductions of this strain were made to the University of Wales Swansea in the 1980s. Several hundred fish, including a number of estrogen-treated phenotypic females, representing more than 10 families were transferred in 1989 to the Philippines where our experiments were carried out.

In this paper the genetic nomenclature for the description of hormone-treated fish described by Mair et al. (1987) is used: the sex symbol refers to the functional reproductive phenotype, with a delta (Δ) prefix denoting functional sex reversal. Thus, a genetic male reversed to female by estrogen treatment is denoted as Δ♀.

Breeding programme

The breeding programme adopted (Fig. 1) has a number of distinct steps.

(1) Feminization of sexually undifferentiated progeny from normal crosses: this has been successfully achieved in this strain (Mair 1988; Scott et al. 1989) and by several authors in a number of other *Oreochromis* species (Scott et al. 1989; Varadaraj 1989; Rosenstein and Hulata 1994).

(2) Identification of sex-reversed females (Δ♀♀ (XY)) by progeny testing: the morphology, behaviour, and karyotypes of sex-reversed females are indistinguishable from those of their genetically female siblings, and thus they can only be identified by progeny testing in crosses with normal XY males. Progeny sex ratios approximating 3:1 are indicative of a maternal XY genotype. Identification by progeny testing of hormonally induced and one naturally occurring Δ♀♀ (XY) was reported by Mair et al. (1991*a*). Five further estrogen-treated Δ♀♀ were identified from among fish transferred to the Philippines.

(3) Crossing of the Δ♀♀ (XY) identified in step 2 with normal males (XY): this should generate progeny including approximately 25% 'YY' males. Each of the five identified Δ♀♀ (XY) was crossed to normal males and the progeny reared for use in progeny testing.

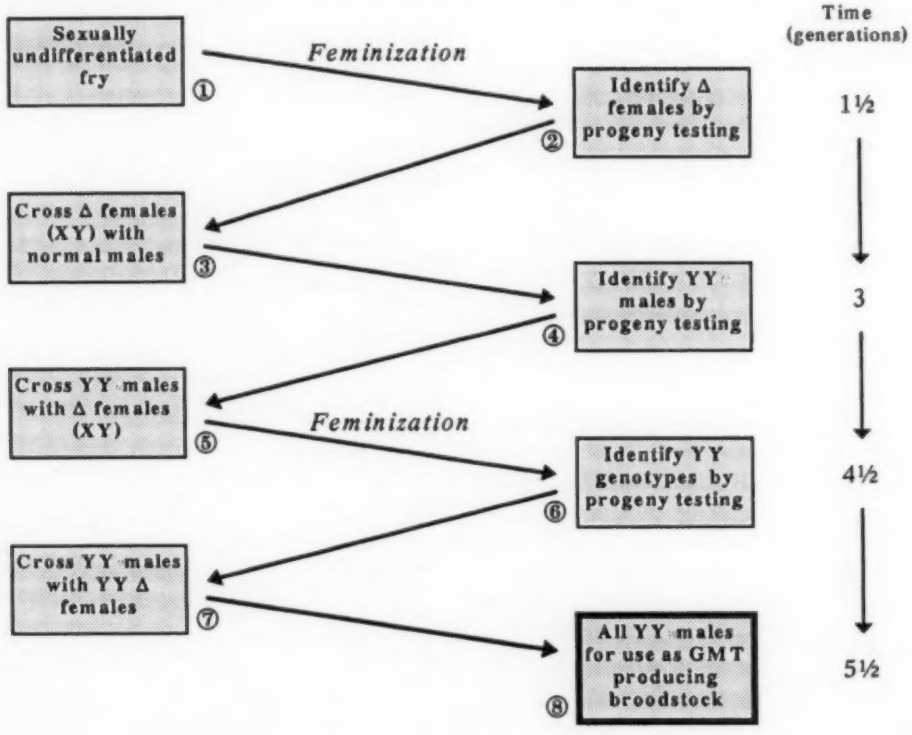
(4) Progeny testing of males to identify YY genotypes: a total of 54 males from these five crosses were progeny tested with normal females. It was hypothesized that 'YY' males would sire only male offspring. Mair et al. (1991*a*) previously reported the identification of four 'YY' males in the progeny of a single naturally occurring Δ♀. Although it is possible to identify 'YY' males at this stage, investment of time and resources in progeny testing would be impractical on a large scale. It is for this reason that it was necessary to complete the succeeding stages of the programme.

(5) Crossing of identified 'YY' males with previously identified Δ♀♀: six Δ♀♀ (five were second-generation Δ♀♀ produced by estrogen treatment of progeny from the initial XY × XY crosses, progeny tested as in step 2) and six 'YY' males were used in nine crosses to generate families with a 1:1 ratio of YY and XY genotypes. The progeny from these crosses were each divided into two batches, one of which was treated with estrogens for feminization.

(6) Progeny testing of estrogen-treated females (Δ♀ (XY) and Δ♀ (YY)) and nontreated males for identification of YY genotypes: it was hypothesized that 50% of all sex-reversed females would be of the YY genotype, assuming no differential feminization of XY and YY genotypes. In the progeny testing of these sex-reversed females with normal XY males, XY females would be expected to produce 3:1 sex ratios, whilst 'YY' females would give all-male or nearly all-male progeny. In practice, it is difficult to discriminate between these two progeny sex ratios statistically for YY × XY crosses that produce

© 1997 NRC Canada

Fig. 1. Schematic diagram of the breeding programme for the large-scale production of YY male broodstock.



some female offspring. Thus, females from all but one of these families were progeny tested using previously identified androgen sex-reversed males (XXΔ♂). Using these males, XY and 'YY' females would produce 1:1 and 1:0 sex ratios, respectively, which are easily separated statistically. A total of 113 sex-reversed females were progeny tested in this way. A total of 158 nontreated males were progeny tested as a control to test the hypothesis that 50% of genotypes would be YY.

(7) Crossing of identified 'YY' males with identified 'YY' females (Δ♀): to produce 'YY' males in large numbers without the need for each to be progeny tested, it was necessary to demonstrate that 'YY' males and 'YY' females could be bred together and would generate sufficient numbers of viable YY male progeny.

(8) Progeny testing of males from YY × YY matings to verify their YY genotype: a total of 61 males from several YY × YY crosses were reared and progeny tested. In addition, some progeny from several YY × YY crosses were feminized to produce future YY male producing broodstock. Twelve of these females were progeny tested using XXΔ♂.

In anticipation of the future production of large numbers of 'YY' males and for the provision of sex-reversed males (Δ♂♂ (XX)) to be used in progeny testing, we also developed all-female lines in the same strain of *O. niloticus*. In this programme, progeny from normal crosses were masculinized by oral application of the androgen 17α-methyltestosterone. These males were progeny tested with normal females and sex-reversed Δ♂♂ (XX) identified on the basis of the hypothesis that they should produce only female progeny. A second generation of masculinisazion enabled the production of large numbers of Δ♂♂ (XX), obviating the need for time-consuming progeny testing. The Δ♂♂ (XX) were viable and produced mean sex ratios of approximately 95% female (G.C. Mair, unpublished data).

Sex reversal by oral application of estrogens and androgens

Functional sex reversal is most easily achieved through oral application of estrogens and androgens incorporated into the feed and administered during the period of sex differentiation, which is known to be fixed by between 14 (Srisakultiew 1993) and 30 days posthatch (Alvendia-Casauay and Carino 1988).

First-feeding fry were collected from incubators following artificial incubation. Fry were transferred at a density of approximately 1000m^{-2} to fine-mesh net cages (1 × 1 × 1 m) suspended in fertilized earthen ponds and fed with a hormone-treated feed, ad libitum, four times daily. Adaptations of protocols for feminization optimized by Rosenstein and Hulata (1994) and Mair and Santiago (1994) were applied as follows: fry were fed with a powdered feed containing 1000 mg diethylstilbestrol (DES)·kg^{-1} for 20 days. Adaptations of commercial protocols were applied for masculinization (Guerrero and Guerrero 1988; Vera Cruz and Mair 1994). Fry were fed a feed containing 17α-methyltestosterone at 40 mg·kg^{-1} for 25 days. Following hormone treatment, fry were transferred to net cages for rearing to sexual maturity.

Progeny testing

For progeny testing, sexually mature fish (150–200 days) were stocked in 1-m^2 fine-mesh cages immersed in earthen ponds to a depth of 60 cm, for spawning, either as single pairs or with one male to three females. Cages were checked every 7 days and eggs or fry removed from incubating females. Spawned males or females were then individually tagged using passive integrated transponder (PIT) electronic tags and stocked communally in concrete tanks. Eggs and fry were incubated artificially up to the completion of yolk sac absorption whereafter they were stocked as sib groups in fine-mesh

© 1997 NRC Canada

Table 1. Summary of progeny testing for potential 'YY' males (crossed to normal XX females) in progeny from XYΔ♀ × XY♂ and XYΔ♀ × YY♂ crosses.

Summary parameter	XYΔ♀ × XY♂	XYΔ♀ × YY♂
No of families from which males were tested	7	3
Number of potential 'YY' males tested	54	158
Number of progeny sexed (mean per family)	3753 (69.5)	10 842 (68.6)
Number of males classified as XY (%)	27 (50.0)	66 (41.8)
Number of males not classified (%)	6 (11.1)	6 (3.8)
Number of males classified as YY (%)	21 (38.9)	86 (54.4)
Expected number of 'YY' males (%)	18 (33.3)	79 (50.0)
χ^2 for observed versus expected number of 'YY' males	0.75 ns	1.24 ns
Range of sex ratios from XY males	37.2–70.0	34.2–70.0
Mean of sex ratios from XY males (±SD)	52.9 (±8.1)	56.6 (±7.9)
Heterogeneity χ^2 for XY males (df)	36.33 (26) ns	90.89 (65) ns
Range of sex ratios from 'YY' males	67.0–100.0	71.7–100.0
Mean of sex ratios from 'YY' males (±SD)	95.31 (±8.4)	96.4 (±7.1)
Heterogeneity χ^2 for 'YY' males (df)	50.21 (20)*	103.90 (78) ns
Total heterogeneity χ^2 for all males (df)	775.64 (53)***	2168.58 (157)***

Note: ns, not significant; *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

cages, for rearing. Fry were reared for approximately 3 months until they attained a mean mass of 3 g, before being killed and sexed using the gonad squash technique of Guerrero and Shelton (1974). In larger families where it was not deemed necessary to sacrifice the entire family, a minimum subsample of 100 fish was taken at random, avoiding size bias, which may have favoured the selection of larger males.

All sex ratios were tested against a 1:1 sex ratio (or 3:1 when testing for XYΔ♀ crossed with normal XY males) using the χ^2 test. A stringent statistical criterion was adopted for the designation of parental genotypes when progeny testing for 'YY' males or females, hypothesized to produce all-male or nearly all-male progeny. A 5% level of probability was not deemed sufficiently stringent to permit the confident designation of genotypes. Thus, potential YY genotypes producing sex ratios, in crosses with XX genotypes, that were not significantly different from 1:1 or only significant at the 5% level ($P > 0.01$) were designated as XY. Only those producing male-skewed sex ratios different from 1:1 at a probability level of 0.1% ($P < 0.001$) were designated YY. This procedure also helped to minimize the chance of making a type I error in the identification of 'YY' males. No genotype was designated for parents of families falling between these two criteria ($0.001 < P < 0.01$).

Proportions of 'YY' males identified in progeny of XY × XY and XY × YY crosses were tested against the 1:2 and 1:1 expectations, respectively, using the χ^2 test.

Results

Five sex-reversed females (Δ♀♀) had previously been identified among DES-treated fish and were used in seven crosses with normal males (XYΔ♀ × XY♂) to produce families including 25% YY genotypes for progeny testing (Mair et al. 1991*a*, unpublished data).

Progeny testing of males from XYΔ♀ × XY♂ crosses

A total of 54 males from seven XYΔ♀ × XY♂ matings were progeny tested in crosses to randomly selected normal females. More than 3500 fingerlings were sexed with an average of 69.5 fingerlings per family (Table 1). This was deemed an adequate sample size to gain an accurate estimate of family sex ratio. Using predefined criteria for allocation of genotypes, 50% of these males were classed as XY, 38.9% as YY, and 11.1% were not classified because their progeny sex ratio was different from 1:1 at $P < 0.01$ but not at $P < 0.001$.

The frequency distribution of family sex ratio is clearly bimodal, those from XY males being normally distributed around a 1:1 sex ratio and a peak at 100% male representing the modal sex ratio for progeny of designated 'YY' males (Fig. 2*a*).

The heterogeneity χ^2 (based on the difference from 1:1 ratios) for the sex ratios of designated XY males was not significant ($\chi^2_{[26]} = 36.33$). However, the sex ratios produced by designated 'YY' males were significantly heterogeneous ($\chi^2_{[20]} = 50.21$) owing to one outlying sex ratio of 67.0% male. If this outlier is removed the χ^2 value becomes nonsignificant ($\chi^2_{[19]} = 15.80$). When all sex ratios are considered together, they are highly heterogeneous ($\chi^2_{[53]} = 775.64$; $P < 0.001$).

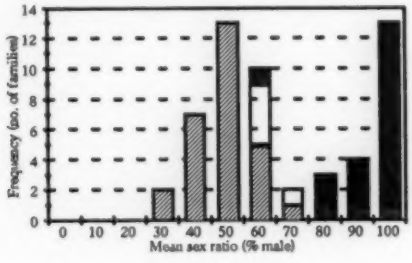
Progeny testing of males from XYΔ♀ × YY♂ crosses

The results from the progeny testing of males from the later crosses of XYΔ♀ × YY♂ followed a similar trend. Of 158 males tested in crosses with normal females, 41.8% were designated as XY, 54.4% as YY, and only 3.8% were unclassified (Table 1). The proportion of 'YY' males was not significantly greater than the predicted 1:1 ratio. Similar range (71.7–100%) and mean (96.4%) proportions of males were observed as with the previously identified 'YY' males. Designated XY males produced sex ratios slightly skewed to male (56.6% male) in this set of progeny tests. The family sex ratio frequency distributions (Fig. 2*b*) showed a pattern similar to that from testing progeny of XYΔ♀ × XY♂ crosses, although, as hypothesized, there was a relatively higher proportion of 'YY' males.

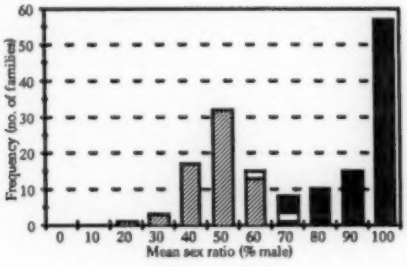
χ^2 heterogeneity values for sex ratios from males designated as XY and as YY were not significant ($\chi^2_{[65]} = 90.89$ and $\chi^2_{[78]} = 103.90$, respectively). All sex ratios considered

© 1997 NRC Canada

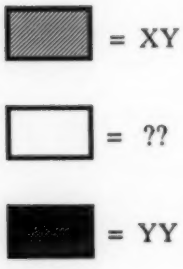
Fig. 2. Histograms showing frequency distributions for family sex ratio in the progeny testing for 'YY' males from XYΔ♀ × XY♂, XYΔ♀ × YY♂, and YYΔ♀ × YY♂ crosses and for 'YY' females in feminized progeny from XYΔ♀ × YY♂ and YYΔ♀ × YY♂ crosses (10% class intervals were used except that the rightmost column represents only 100% males).

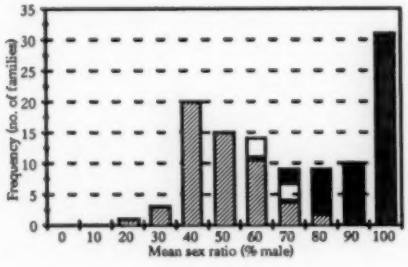


(a) Males from XY x XY crosses

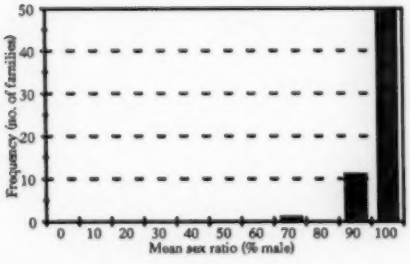


(b) Males from XY x YY crosses

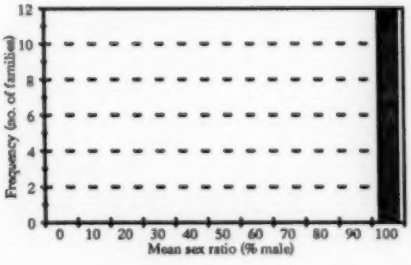




(c) Females from XY x YY crosses



(d) Males from YY x YY crosses



(e) Females from YY x YY crosses

together were highly heterogeneous ($\chi^2_{[157]}$ = 2168.58; $P < 0.001$).

Progeny testing of DES-treated females from XYΔ♀ × YY♂ crosses

Females were selected for progeny testing at random from nine DES-treated families of XYΔ♀ × YY♂ crosses (see Table 2). A total of 113 females were tested, the majority (105) in crosses with sex-reversed XXΔ♂ to facilitate statistical discrimination of XY and YY genotypes. No 'YY' genotypes were designated from the eight progeny tests of females from family 1, which were tested with XY males. This is lower ($P < 0.01$) than the expected proportion and is possibly a consequence of the difficulty in designating genotypes associated with the 3:1 sex ratios expected from tests of XYΔ♀. Overall, 50.4% of the 113 females were classed as XY, 45.1% were classed as YY, and only 4.5% were unclassified. Other than family 1, no families had proportions of 'YY' females differing ($P > 0.05$) from the expected 1:1, although it should be noted that only a small number of females were tested in each

© 1997 NRC Canada

Table 2. Results from progeny testing of females resulting from feminization of progeny from XYΔ♀ × YY♂ matings.

	Family number									
	1	2	3	4	5	6	7	8	9	Total
Source cross (XYΔ♀ × YY♂)[a]	1 × 1	1 × 4	2 × 2	2 × 3	3 × 3	4 × 2	5 × 3	5 × 5	6 × 6	6Δ♀ × 6♂
Sex ratio following DES treatment	16:24	16:15	5:27	0:11	0:14	2:3	1:64	—	—	40:158
Male genotype used in progeny test	XY	XX	XX	XX	XX	XX	XX	XX	XX	—
Number of females tested	8	15	10	4	5	3	50	12	6	113
Number of progeny sexed	420	732	423	242	238	124	2450	589	373	5591
Number of females classified as XY	8	8	7	1	0	2	23	3	5	57
Number of females not classified	0	1	0	0	0	1	1	1	1	5
Number of females classified as YY	0	6	3	3	5	0	26	8	0	51
Expected proportion of YYΔ♀[b]	4	7.5	5	2	2.5	1.5	25	6	3	56.5
Exact probability for binomial test	0.008***	0.790 ns	0.344 ns	0.625 ns	0.063 ns	0.500 ns	0.775 ns	0.228 ns	0.062 ns	0.54
Range of sex ratios from XYΔ♀♀[c]	63.6–85.0	35.0–65.0	37.0–61.0	n/a	n/a	39.5–48.9	42.4–65.0	44.4–54.7	27.0–66.0	35.0–85.0
Mean sex ratio from XYΔ♀♀[c]	75.1	50.4	50.9	48.0	n/a	44.2	53.6	50.4	47.3	52.5
Range of sex ratios from YYΔ♀♀[c]	n/a	92.0–100.0	97.4–100.0	85.0–100.0	n/a	n/a	74.0–100.0	80.5–100.0	n/a	74.0–100.0
Mean sex ratio from YYΔ♀♀[c]	n/a	98.7	99.2	94.4	100.0	n/a	94.0	97.4	n/a	97.3

[a]Numbers represent different individual males and females.
[b]Expected proportion based on hypothesis of 1:1 ratio of XY to YY genotypes, with equal feminization rates.
[c]Sex ratios given as percentage males.

family. The overall range (74.0–100.0% male) and mean (97.3% male) of the sex ratios produced by 'YY' females were in accordance with those produced by 'YY' males.

The sex ratio frequency distribution for the progeny testing of DES-treated females (Fig. 2*c*) closely resembled that for the males (Fig. 2*b*).

Progeny testing of males and females from YYΔ♀ × YY♂ crosses

Following successful identification of both 'YY' males and 'YY' females, a number of YYΔ♀ × YY♂ crosses were attempted. Several spawnings were obtained and progeny were divided into two, with one group receiving DES treatment. Table 3 shows the summary of the results from the progeny testing of 61 males (with normal XX♀♀) from four such crosses and 12 sex-reversed females (with sex-reversed XXΔ♀♀) from three crosses. Following the sexing of over 4300 progeny, all tested males and females were designated as YY genotypes. All 12 females produced 100% male progeny (Fig. 2*e*) and the 'YY' males produced a mean sex ratio of 98.9% male (Fig. 2*d*). Sex ratios from these 'YY' males were highly homogeneous ($\chi^2_{[60]} = 11.27$; $P > 0.99$).

Summary of sex ratios produced by XY and YY genotypes

In the progeny testing of males from XY × XY and XY × YY crosses, designated XY males produced ratios slightly skewed to male. The total sex ratio from all 93 XY males was 52.4% male, highly significantly greater than the predicted 1:1 ratio ($\chi^2_{[1]} = 13.14$; $P < 0.001$).

Over all progeny tests, excluding those males from YY × YY crosses, fish designated as YY genotypes produced sex ratios ranging from 67.0 to 100% male with a mean of 95.63 ± 7.87% male. It was noted that the 'YY' males from the YY × YY cross gave a higher mean sex ratio of 98.90 ± 3.1% male. The 'YY' males and females used in these YY × YY crosses were selected on the basis of the sex ratio of their initial progeny tests (all had produced 100% male progeny). These results therefore indicate a possible response to selection against the occurrence of small proportions of females.

To test the hypothesis of a genetic basis for the occurrence of small proportions of females in the putative all-male progeny of 'YY' males, a number of males were selected for repeat progeny tests. Twenty-six males that had produced initial progeny test sex ratios of >95% male (mean ♂ 99.67%) were selected and repeat mated one or more times (sex ratios were pooled for males repeat mated). The mean sex ratio of these repeat progeny tests was 99.66% male, higher than the 95.63% mean sex ratio for all 'YY' males, this difference being almost significant ($P = 0.0502$) as determined by a Mann–Whitney nonparametric test.

Discussion

Sex determination in *O. niloticus*

Our results provide unequivocal evidence for an underlying basic monofactorial mechanism of sex determination in this strain of *O. niloticus* and also for the viability of novel 'YY' males. The results from the progeny testing for 'YY' males from XY × XY crosses, in which 'YY' males were present in expected proportions, indicate that the 'YY' males have viability and fertility equivalent to normal males. A similar

© 1997 NRC Canada

Table 3. Summary of progeny testing for putative 'YY' males (crossed to normal XX♀♀) and 'YY' females (crossed to sex-reversed XXΔ♂♂) in progeny from YYΔ♀ × YY♂ crosses.

Summary parameter	'YY' males	'YY' females
Number of families from which males were tested	4	3
Number of putative YY genotypes tested	61	12
Number of progeny sexed (mean per family)	3723 (61.0)	652 (54.3)
Number of males classed as YY (%)	61 (100.0)	12 (100.0)
Expected number of 'YY' males (%)	61 (100.0)	12 (100.0)
χ^2 value of observed versus expected number of 'YY' males	0.0	0.0
Range of sex ratios from 'YY' males	79.5–100.0	100.0
Mean of sex ratios from 'YY' males (±SD)	98.9 (±3.1)	100 (±0)

pattern was observed with the progeny testing of males from the XY × YY crosses, with the number of 'YY' males not differing significantly from that expected on the basis of a theory of segregation of a single male-determining facto. (gene or supergene).

The distinct bimodal sex ratio distribution of the families from these two sets of progeny tests and the relatively low proportion of unassigned genotypes support the validity of the arbitrary criterion used in the designation of genotype. This is further supported by the highly significant heterogeneity of the sex ratios of all families, considered together, compared with the relative homogeneity of sex ratios from the groups of males designated separately as XY and YY genotypes. Progeny sex ratios of males designated as normal XY males varied from 34.2 to 70.0% but with a mean close to 50% male, with a slight but significant overall excess of males. These sex ratios thus appear to be normally distributed about the expected 1:1 ratio of males to females. This distribution is similar to, but slightly more heterogeneous than, that observed for the same Egyptian strain of *O. niloticus* by Mair et al. (1991*a*) and slightly more homogenous than those observed by Shelton et al. (1983) in an Ivory Coast strain of *O. niloticus*. Lester et al. (1989) observed considerably more heterogeneity in the sex ratios of families collected from a mix of strains, some of which were known to be introgressed with *O. mossambicus* (Macaranas et al. 1986). The authors interpreted these high levels of heterogeneity for sex ratio as evidence for polyfactorial sex determination in this species, and it is possible that some strains of "*O. niloticus*" may have such polyfactorial mechanisms, especially when introgressed with other species.

The sex ratios produced by the males that were designated as YY genotypes were not quite in accordance with those predicted by the hypothesis of simple monofactorial sex determination, many of them being somewhat lower than the expected 100% male. This observation is in accordance with the results of Mair et al. (1991*a*) who observed a single female in the progeny of one of four 'YY' males progeny tested. Scott et al. (1989), on the other hand, observed no females in the sexing of 285 progeny of a single YY male crossed to 10 separate females. Similarly, Varadaraj and Pandian (1989) observed no females among the progeny of eight 'YY' males in *O. mossambicus*. It is probable that the greater sample sizes and the larger number of fish tested are responsible for the more numerous observations of females in progeny of 'YY' males in this study. The overall sex ratio from 'YY' males was greater than 95% male, with the majority of ratios being 100%. There was no apparent trend in the occurrence of these aberrant sex ratios that would indicate the segregation of a single autosomal sex-modifying locus as postulated for *O. niloticus* by Hussain et al. (1994) and demonstrated for *O. aureus* by Mair et al. (1991*b*).

The increase in the overall proportions of males in the progeny testing of YY genotypes derived from crosses of selected (on the basis of the 100% male sex ratios in initial progeny tests) 'YY' males indicates some form of response to this selection and thus a genetic basis to the occurrence of these aberrant females. Similarly the higher than average proportions of males in repeat matings of "selected" (i.e., produced >96% male sex ratios in initial progeny tests) 'YY' males provide further support for this hypothesis of a genetic basis. It cannot be discounted, however, that the overall mean sex ratio of all fish derived from males designated as YY was lower than that from the repeated matings of selected 'YY' males because of the erroneous inclusion of one or more sex ratios from incorrectly designated XY males.

In a recent study in the same strain, Capili (1995) demonstrated that these "rare" females, arising in progenies of 'YY' males, conform to the expected XY genotype in progeny tests with known genotypes. In a separate study, Capili (1995) also noted the existence of significant paternal and maternal effects on the occurrence of rare females in the progeny of 'YY' males. It seems likely that a series of autosomal sex-modifying genes, acting as a threshold trait, may be responsible for the occurrence of these rare females and that these autosomal genes could be selected against to further increase the proportion of males in the progeny of 'YY' males. It is also likely, however, that there is a temperature effect on sex differentiation. This has been demonstrated in the occurrence of males in the putative all-female progeny of sex-reversed XXΔ♂♂ (Baroiller et al. 1995; J.S. Abucay, unpublished data) and may also play a role in the occurrence of these rare females. The seasonally higher temperatures encountered in the Philippines, which are more extreme than those in the natural range of this species, may also be a factor in the slight excesses of males in the progeny of normal XY males observed in this and other studies.

© 1997 NRC Canada

Potential for application in aquaculture

The successful feminization of the YY genotype is the first to be reported in tilapia and is a vital step in the development of the YY technology on a large scale, as it makes possible the production of 'YY' males without the need for time-consuming progeny testing. In their work on *O. mossambicus* in the 1970s, Yang et al. (1980) produced up to 600 'YY' males in XY × YY matings, although it is not clear whether these were identified in progeny testing. There are no reports of 'YY' males being used commercially for the production of all-male *O. mossambicus*, and it seems likely that these 'YY' males were not produced on a commercial scale, possibly owing to a failure to produce 'YY' females by feminization. We hypothesized that 'YY' males may be more resistant to feminization. This was investigated in a study by Abucay and Mair (1997) in which feminized females and nonfeminized males from DES-treated progeny of XY × YY crosses were progeny tested; evidence was presented for differential feminization of XY and YY genotypes in one of three families from which males were tested. Our data do not support the hypothesis of differential feminization of XY and YY genotypes, although it is possible that this occurred in two of the nine families from which males were tested.

The successful mating of 'YY' males and 'YY' females (all the progeny of which were male) and the confirmation of the YY genotype of the males and DES-treated females from these crosses establish the potential for development of this technology on a commercial scale. 'YY' males and females can now be produced in large numbers for use as broodstock to mass produce 'YY' males. In turn, these can be crossed with females, which can also be mass produced from XX♀ × XXΔ♂ crosses, to generate commercially applicable numbers of genetically male tilapia (GMT) for culture.

The progeny of the 'YY' males (the term GMT distinguishes them from sex-reversed male tilapia) have been comprehensively evaluated in on-station and on-farm trials. Results from on-station trials indicate that GMT have very considerable benefits under culture, significantly increasing yields by up to 58%, compared with mixed-sex tilapia of the same strain (Mair et al. 1995). Yields of GMT are also consistently greater than those for sex-reversed male tilapia. In addition to the negligible recruitment in GMT populations, they have the further advantages of more uniform harvest size distribution, higher survival, and better food conversion ratios.

There are several important comparative advantages and disadvantages of the YY male technology as a means for production of monosex male tilapia, as compared with other commonly used alternatives such as manual sexing, hybridization, and sex reversal. The technique can be considered environmentally friendly, as no hormones are applied to fish that are consumed and, overall, hormone application to broodstock is very low. Species–strain purity is maintained and the fish produced for culture are normal genetic males. Although the development process is time consuming and labour intensive, once developed the production of monosex males can be maintained through occasional feminization of YY genotypes. Provided that broodstock purity can be maintained, the technology can be applied (at the level of spawning 'YY' males with normal females) in existing hatchery systems without any special facilities or labour requirements. Discounting the initial development costs, additional costs for application of this technology at the hatchery level would be minimal, while the potential economic advantages to growers have been demonstrated to be very considerable.

Attempts are being made to extend this technology to the tilapia culture industry in the Philippines as a unique example of application of genetic manipulation technology in developing-country aquaculture.

Acknowledgments

The research reported here was conducted over 6 years, from 1989 to 1995, at the Freshwater Aquaculture Center, Central Luzon State University, Philippines. We acknowledge the assistance of Josie Capili and Eric Roderick and the dedication of the Genetic Manipulations for Improved Tilapia (GMIT) project staff over the years. We are grateful for the enthusiastic cooperation of the members of the Freshwater Aquaculture Center. This research was funded by a grant from the Fish Genetics Research Programme of the Overseas Development Administration (R. 4803) to G.C.M. and D.O.F.S.

References

Abucay, J.S., and Mair, G.C. 1997. Methods of identifying males with YY genotype in *Oreochromis niloticus* L. *In* Proceedings of the Second AADCP International Workshop on Genetics in Aquaculture and Fisheries Management, Phuket, Thailand, Nov. 7–11, 1994.

Alvendia-Casauay, A., and Carino, V.S. 1988. Gonadal sex differentiation in *Oreochromis niloticus*. *In* ICLARM Conference Proceedings, 15: The Second International Symposium on Tilapia in Aquaculture. *Edited by* R.S.V. Pullin, T. Bhukaswan, K. Tonguthai, and J.L. Maclean. Department of Fisheries, Thailand, and International Center for Living Aquatic Resources Management, Bangkok, Thailand, and Manila, Philippines. pp. 121–124.

Baroiller, J.-F., and Jalabert, B. 1989. Contribution of research in reproductive physiology to the culture of tilapias. Aquat. Living Resour. **2**: 105–116.

Baroiller, J.-F., Chourrout, D., Fostier, A. and Jalabert, B. 1995. Temperature and sex chromosomes govern sex-ratios of the mouthbrooding cichlid fish *Oreochromis niloticus*. J. Exp. Zool. **271**: 1–8.

Bye, V.J., and Lincoln, R.F. 1986. Commercial methods for the control of sexual maturation in rainbow trout (*Salmo gairdneri* R.). Aquaculture, **57**: 299–309.

Capili, J.B. 1995. Growth and sex determination in the Nile tilapia (*Oreochromis niloticus* L.). Ph.D. thesis, University of Wales, Swansea.

Food and Agriculture Organization of the United Nations. 1995. Aquaculture production statistics 1984–1993. FAO Fish. Circ. No. 815 Rev. 7.

Guerrero, R.D., III, and Guerrero, L.A. 1988. Feasibility of commercial production of sex-reversed Nile tilapia fingerlings in the Philippines. *In* ICLARM Conference Proceedings, 15: The Second International Symposium on Tilapia in Aquaculture. *Edited by* R.S.V. Pullin, T. Bhukaswan, K. Tonguthai, and J.L. Maclean. Department of Fisheries, Thailand and International Center for Living Aquatic Resources Management, Bangkok, Thailand, and Manila, Philippines. pp. 183–186.

Guerrero, R.D., III, and Shelton, W.L. 1974. An aceto-carmine squash method of sexing juvenile fishes. Prog. Fish-Cult. **36**: 56.

Hammerman, I.S., and Avtalion, R.R. 1979. Sex determination in *Sarotherodon* (*Tilapia*). Theor. Appl. Genet. **55**: 177–187.

Hussain, M.G. 1992. Genetic manipulations studies in *Oreochromis niloticus* L. Ph.D. thesis, Institute of Aquaculture, Stirling, Scotland.

© 1997 NRC Canada

Hussain, M.G., McAndrew, B.J., Penman, D.J., and Sodsuk, P. 1994. Estimate gene-centromere recombination frequencies in gynogenetic diploids of *Oreochromis niloticus* (L.) using allozymes, skin colour and a putative sex-determination locus (*SDL-2*). *In* Genetics and evolution of aquatic organisms. *Edited by* A.R. Beaumont. Chapman and Hall, London, U.K. pp. 502–508.

Lester, L.J., Lawson, K.S., Abella, T.A., and Palada, M.S. 1989. Estimated heritability of sex ratio and sexual dimorphism in tilapia. Aquacult. Fish. Manage. **20**: 369–380.

Macaranas, J.M., Taniguchi, N., Pante, M.J.R., Capili, J.B., and Pullin, R.S.V. 1986. Electrophoretic evidence for extensive hybrid gene introgression into commercial *Oreochromis niloticus* (L.) stocks in the Philippines. Aquacult. Fish. Manage. **17**: 249–258.

Mair, G.C. 1988. Studies on sex determining mechanisms in *Oreochromis* species. Ph.D. thesis, University of Wales, Swansea.

Mair, G.C., and Little, D.C. 1991. Population control in farmed tilapias. NAGA, **14**: 8–13.

Mair, G.C., and Santiago, L.P. 1994. Feminization of Nile tilapia *Oreochromis niloticus* (L.) by oral application of diethylstilbestrol (DES). *In* The Third Asian Fisheries Forum. *Edited by* L.M. Chou, A.D. Munro, T.J. Lam, T.W. Chen, L.K.K. Cheong, J.K. Ding, K.K. Hooi, H.W. Khoo, V.P.E. Phang, K.F. Shim, and C.H. Tan. Asian Fisheries Society, Manila, Philippines. pp. 94–97.

Mair, G.C., Penman, D.J., Scott, A., Skibinski, D.O.F., and Beardmore, J.A. 1987. Hormonal sex-reversal and the mechanisms of sex determination in *Oreochromis*. *In* Proceedings of the World Symposium on Selection, Hybridization, and Genetic Engineering in Aquaculture, Bordeaux, 27–30 May 1986. Vol. II. *Edited by* K. Tiews. H. Heenemann Verlagsgesellschaft mbH, Berlin. pp. 301–312.

Mair, G.C., Beardmore, J.A., and Skibinski, D.O.F. 1990. Experimental evidence for environmental sex determination in *Oreochromis* species. *In* The Second Asian Fisheries Forum. *Edited by* R. Hirano and I. Hanyu. Asian Fisheries Society, Manila, Philippines. pp. 555–558.

Mair, G.C., Scott, A., Penman, D.J., Beardmore, J.A., and Skibinski, D.O.F. 1991*a*. Sex determination in the genus *Oreochromis*. I. Sex reversal, gynogenesis, and triploidy in *O. niloticus* L. Theor. Appl. Genet. **82**: 144–152.

Mair, G.C., Scott, A., Penman, D.J., Skibinski, D.O.F., and Beardmore, J.A.. 1991*b*. Sex determination in the genus *Oreochromis*. II. Sex reversal, hybridisation, gynogenesis and triploidy in *O. aureus* Steindachner. Theor. Appl. Genet. **82**: 153–160.

Mair, G.C., Abucay, J.S., Beardmore, J.A., and Skibinski, D.O.F. 1995. Growth performance trials of genetically male tilapia (GMT) derived from 'YY' males in *Oreochromis niloticus* L.: on-station comparisons with mixed sex and sex reversed male populations. Aquaculture, **137**: 313–322.

Pullin, R.S.V., and Maclean, J.L. 1992. Analysis of research for the development of Tilapia farming: an interdisciplinary approach is lacking. Neth. J. Zool. **42**: 512–522.

Rosenstein, S., and Hulata, G. 1994. Sex reversal in the genus *Oreochromis*: optimization of feminization protocol. Aquacult. Fish. Manage. **25**: 329–339.

Scott, A.G., Penman, D.J., Beardmore, J.A., and Skibinski, D.O.F. 1989. The 'YY' supermale in *Oreochromis niloticus* (L.) and its potential in aquaculture. Aquaculture, **78**: 237–251.

Shelton, W.L., Meriwether, F.H., Semmens, K.J., and Calhoun, W.E. 1983. Progeny sex ratios from intraspecific pair spawnings of *Tilapia aurea* and *T. nilotica*. *In* Proceedings of the International Symposium on Tilapia in Aquaculture, Nazareth, May 1983. *Edited by* L. Fishelson and Z. Yaron. Tel Aviv University, Israel. pp. 270–280.

Srisakultiew, P. 1993. Studies on the reproductive biology of *Oreochromis niloticus* L. Ph.D. thesis, University of Stirling, Scotland.

Trombka, D., and Avtalion, R.R. 1993. Sex determination in tilapia. a review. Isr. J. Aquacult. Bamidgeh, **45**: 26–37.

Varadaraj, K. 1989. Feminization of *Oreochromis mossambicus* by the administration of diethylstilbestrol. Aquaculture, **80**: 337–341.

Varadaraj, K., and Pandian, T.J. 1989. First report on production of supermale tilapia by integrating endocrine sex reversal with gynogenetic technique. Curr. Sci. **58**: 434–441.

Vera Cruz, E.M., and Mair, G.C. 1994. Conditions for effective androgen sex reversal in *Oreochromis niloticus* (L.). Aquaculture, **122**: 237–248.

Wohlfarth, G.W. 1994. The unexploited potential of tilapia hybrids in aquaculture. Aquacult. Fish. Manage. **25**: 781–788.

Yang, Y., Zhang, Z., Lin, K., Wei, Y., Huang, E., Gao, A., Xu, Z., Ke, S., and Wei, J. 1980. Use of three line combination for production of genetic all-male *Tilapia mossambica*. Acta Sci. Sin. **7**: 241–246.

© 1997 NRC Canada

Size- and species-dependent variability in consumption and growth rates of larvae and juveniles of three freshwater fishes

Benjamin H. Letcher, James A. Rice, Larry B. Crowder, and Fred P. Binkowski

Abstract: We tested the hypothesis of no species differences in consumption and growth rates using three species of Lake Michigan fish (yellow perch (*Perca flavescens*), bloater (*Coregonus hoyi*), and alewife (*Alosa pseudoharengus*)), that hatch over a wide range of sizes (3.5–9.5 mm). Ingestion and growth rates were significantly different among the three species for 10- and 20-mm fish, although differences were weaker for 10-mm growth rates. Incorporating these consumption and growth rate estimates and their parameter errors into a simulation model revealed that differences among species in survival could be detected with as few as 3 samples (model runs) for 20-mm fish at both low and high prey density and for 10-mm fish at low prey density, but that 23 samples would be required to detect differences for 10-mm fish at high prey density. Compared with our 8-h estimates, previous short-term (15–30 min) consumption rate estimates for the same three species overestimated consumption rates for the 20-mm fish but underestimated them for the 10-mm fish. Our results suggest that differences in growth and ingestion among the three species are likely even when fish are 10 mm, but that we may not always be able to detect differences in survival of smaller fish.

Résumé : Nous avons vérifié l'hypothèse qu'il n'y avait pas de différences entre les espèces en ce qui a trait aux taux de consommation et de croissance chez trois espèces de poisson du lac Michigan, la perchaude (*Perca flavescens*), le cisco de fumage (*Coregonus hoyi*), et le gaspareau (*Alosa pseudoharengus*), qui, à l'éclosion, présentent une gamme varié de tailles (3,5 à 9,5 mm). Les taux d'ingestion et de croissance différaient de manière statistiquement significative entre les trois espèces pour les poissons de 10 et 20 mm, bien que les différences étaient moins marquées dans le cas des taux de croissance chez les poissons de 10 mm. L'incorporation de ces valeurs estimées des taux de consommation et de croissance ainsi que des erreurs sur les paramétriques qui les accompagnent dans un modèle a révélé que les différences de survie entre les espèces pouvait être déterminées avec seulement trois échantillons (applications du modèle) pour les poissons de 20 mm à densité de proies élevée et faible et pour les poissons de 10 mm à densité de proies faible, mais que 23 échantillons seraient nécessaires pour déceler les différences pour les poissons de 10 mm à densité de proies élevée. Comparativement à nos valeurs estimées de 8 h, les valeurs estimées antérieures de la consommation à court terme (15–30 min) pour les trois mêmes espèces entraînaient une surestimation des taux de consommation chez les poissons de 20 mm, mais une sous-estimation de ces derniers chez les poissons de 10 mm. Nos résultats laissent entendre que les différences dans la croissance et la consommation chez les trois espèces sont vraisemblablement égales lorsque les poissons ont une longueur de 10 mm, mais que nous ne pourrions pas toujours être en mesure de détecter les différences de survie chez les poissons plus petits. [Traduit par la Rédaction]

Introduction

Small differences in consumption and growth rates can have a substantial impact on survival of larval and juvenile fishes (Houde 1987; Beyer 1989) because many of the factors regulating mortality are size dependent (Blaxter 1986; Anderson 1988; Miller et al. 1988). Because year-class strength and eventually recruitment often are determined by mortality rates while fish are in the larval and juvenile stages (Sissenwine 1984), growth variability during early life can have important population-level consequences (Rice et al. 1993).

Because of the importance of size during the early ontogeny of fishes, researchers have begun to ask recently whether size alone can be used to predict performance of young fish across species. In support of this view, Miller et al. (1988) found that variability in many of the behavioral and energetic processes that may influence survival could be explained by fish length. In further studies, size explained more of the variance in visual acuity and functional responses of larvae and juveniles of three Lake Michigan fishes than did species (Miller et al. 1992, 1993). A size-based approach is attractive because it suggests that generality may exist across species so that we could reasonably predict performance on the basis of size, and that we may be able to use size-based

Received August 31, 1995. Accepted July 16, 1996.
J13061

B.H. Letcher,[1] **J.A. Rice, and L.B. Crowder.**[2] Department of Zoology, Box 7617, North Carolina State University, Raleigh, NC 27695-7617, U.S.A.
F.P. Binkowski. Center for Great Lakes Studies, University of Wisconsin-Milwaukee, 600 East Greenfield Avenue, Milwaukee, WI 53204, U.S.A.

[1] Author to whom all correspondence should be sent at the following address: USGS/Biological Resources Division, S.O. Conte Anadromous Fish Research Center, One Migratory Way, P.O. Box 796, Turners Falls, MA 01376, U.S.A. e-mail: bletcher@external.umass.edu
[2] Present address: Duke University School of the Environment, Marine Laboratory, 135 Duke Road, Beaufort, NC 28516-9721, U.S.A.

© 1997 NRC Canada

estimates as an initial prediction for less well studied species (Miller et al. 1992). Despite the importance of growth during early life, little is known about the relative effects of species and size on growth rate (but see Houde and Schekter 1981). If fish of the same size, but from different species, have very different growth rates under identical conditions, then it will be important to estimate growth for each species individually to predict consumption and growth rates. If, on the other hand, growth rates of different species are similar and scale with body size, specification of size alone should provide adequate growth predictions.

Estimating the variability in growth and consumption among species is also important because it should help in sorting out sources of variability in survival. The potentially large effects of growth rate variability on survival and recruitment (Houde 1987; Rice et al. 1993) suggest that species differences in growth, interacting with size-dependent sources of mortality, could play a part in recruitment variability among species. Species differences, however, will only influence survival if the variability within species is substantially less than the variability among species. Estimating the relative importance of within- and among-species variability in growth rates under controlled conditions may provide data that can be used to determine whether species differences are sufficiently large to contribute to among-species variability in survival.

The goal in this paper was to test the null hypothesis of no species differences in consumption and growth rates for three Lake Michigan fishes, alewife (*Alosa pseudoharengus*), bloater (*Coregonus hoyi*), and yellow perch (*Perca flavescens*), at common lengths over a range of prey densities (0–1000·L^{-1}). We chose these species because they hatch over a broad range of sizes (3.5–9.5 mm) and because culture techniques are well established (Miller et al. 1992). We conducted the experiments when the fish were identical in length, but at these lengths they varied in age and mass. In the analyses, we controlled for mass by expressing feeding and growth rates as proportions of body mass, but we could not control for age differences. In addition to testing for species effects, we also compared consumption rate predictions from short-term (15–30 min) feeding experiments (Miller et al. 1992) using the same sizes and species with the 8-h estimates obtained as part of this study. With this analysis, we assessed the applicability of short-term functional response estimates to longer-term predictions. Finally, we explored the population-level consequences of our consumption and growth rate variability estimates with a survival simulation model.

Materials and methods

Experiments were conducted at the Center for Great Lakes Studies, Milwaukee, Wisconsin, during the spring and summer of 1991 and 1992. Ripe females of each of the three species were caught in the field and the embryos were fertilized (alewife, Milwaukee Harbor; bloater, Lake Michigan; yellow perch, Green Bay, Lake Michigan). Eggs were incubated in the laboratory until hatch. On hatching the fish were transferred to 700-L fiberglass tanks, where they were raised until the beginning of the trials (10 or 20 mm). Water temperatures in the tanks were increased slowly (<1°C·day^{-1}) to 22°C before the 10-mm trials. During rearing, we fed the larvae a mixed assemblage of cultured plankton (63–425 μm), *Artemia* spp. nauplii, commercial fish food (Biodiet starter Nos. 2 and 3), and a ground beef liver mixture (125–450 μm) and maintained a 12 h light : 12 h dark cycle. Two days before each experiment the fish were fed *Artemia* spp. nauplii exclusively.

We analyzed consumption and growth rates of each species at initial body sizes of 10 and 20 mm in 7-day trials. Fish were exposed to five prey density treatments (0, 10, 50, 250, and 1000 *Artemia* spp. nauplii/L), each replicated three times. One day before the beginning of each trial, we selected fish of the appropriate size (10 or 20 mm) from the rearing tanks and transferred them to the experimental tanks. The tanks were inverted 22-L polycarbonate spring-water bottles that each contained 9 L of water maintained at 22 ± 1°C by a water bath. Ten fish were placed in each tank in the 10-mm size experiments and five fish were added to each tank in the 20-mm experiments. Ten to 25 fish were also subsampled from the 700-L stock tanks for initial length and mass measurements.

Artemia nauplii tend to sink in fresh water. We used a nauplii resuspension system (Letcher 1994) to maintain an even distribution of nauplii in the tanks during the course of each 8-h feeding period. Blowing air across the surface of the water along the edge of an inverted spring-water bottle created a weak circulation that pushed sinking nauplii to the center of the bottle where they were resuspended by rising air and water. With this method, it appeared that we achieved an even distribution of nauplii with minimal disturbance to the fish.

Because feeding rate was one of our response variables and may be a function of prey density, species, or fish size, we needed to keep densities, an independent variable, as constant as possible. During trials, we replenished prey densities every 2 h after estimating prey depletion during the previous 2 h by counting the prey with an image analyzer (details below). This approach was sensitive to changes in fish consumption rates, provided good estimates of daily feeding, and replenished *Artemia* spp., which tend to die in fresh water after 6 h. The number of prey eaten in each tank was estimated every 2 h by subtracting the estimated number of prey in the tank after 2 h of feeding from the estimated number at the beginning of each 2-h period. Prey were sampled in duplicate at the end of each 2-h period. To calculate the number of nauplii in each tank, the volume removed was measured with a graduated cylinder (average volume = 0.26 L) and the count was divided by that volume to yield number per litre. To maintain prey densities at average nominal levels, densities in each tank were corrected at the end of each 2-h feeding period using the following equation:

$$\text{(1)} \qquad \text{number added} = \frac{\text{ATE}}{2} + (\text{NOM} - \text{SAMPLE})$$

where number added equaled the number of nauplii to add at the beginning of the next 2-h period, ATE equaled the number eaten in the previous 2-h period, NOM was the nominal density ((9 L)$^{-1}$), and SAMPLE was the number sampled ((9 L)$^{-1}$) at the end of the 2-h feeding period. Because fish in the 20-mm experiments could totally deplete prey in less than 2 h in the 10 and 50·L^{-1} treatments, feedings were supplemented 1 h into each feeding period (after counting with the image analyzer) for those two treatments.

We added the correct number of nauplii by counting individual nauplii with an image analyzer (10 and 50·L^{-1} treatments) or by adding the appropriate volume of known-density nauplii stock (250 and 1000·L^{-1} treatments), using PcVisions+ frame grabber board with Java software (Jandel Scientific). Images for analysis were obtained with a video camera (Panasonic WV-S050 system with WV-5000 camera) aimed at the counting bowl (details in Letcher 1994). The density of the nauplii stock was established each day from the mean of 11 samples (0.25–1 mL each). The coefficient of variation of the samples was never above 4% and was usually between 1 and 3%. There was a good relationship between actual counts (counted under a microscope) and image analyzer counts up

© 1997 NRC Canada

to about 400 nauplii (actual = 1.2 + 0.99 × image analyzer, r^2 = 0.98, n = 8).

To count the uneaten nauplii at the end of each day of feeding (8 h total) and to replace water, 7 L of water was siphoned from each tank and the nauplii were collected on a sieve. We assumed that any nauplii remaining in the tanks were eaten during the next day. The collected nauplii were counted (by eye under a microscope because of contamination with feces and detritus) and were subtracted from the total number added that day (minus the estimated number of nauplii remaining after siphoning) to yield a daily estimate of the number of nauplii eaten.

Individual fish were measured in length (total length) and mass (wet and dry) for estimates of growth. Lengths were taken with an ocular micrometer (±0.1 mm) in a microscope for fish <25 mm and with ruler for fish >25 mm (±0.2 mm). After removing excess water with absorbent paper, we measured individual wet masses on a Mettler balance (±0.01 mg) and weighed dried fish (>24 h at 60°C) on a Cahn microbalance (±1 μg). Because there was substantial mortality of fish in the 0·L^{-1} treatment before the end of the trials, they were removed and measured once 50% of the fish in each tank had died.

Data analysis

We could not standardize for all of the differences among species (age, mass, length, developmental stage) in a single experiment; we chose to conduct experiments at identical lengths. We compared among species of a particular length to ask whether there were length-dependent species differences. If the variability within species was similar to that among species and there were no differences among species at a given length, we could conclude that length alone was sufficient to describe feeding or growth rates. If, conversely, there were species differences at a given length, we needed to test for the effect of the other measure of size: mass (which differed among species at a given length). We did this by expressing the dependent variables (consumption and growth rate) as a proportion of mean fish body mass during a trial. If there were no differences among species after we removed the effect of mass, we could conclude that we were unable to reject the null hypothesis of no species differences and that size alone (length and (or) mass) reasonably predicted feeding and growth rates.

We calculated actual prey densities for each tank as the average prey density during the 7 days of a trial. Average daily prey density equaled the mean of the eight daily prey density estimates (data points in Fig. 1). To standardize for mass differences among the species during each experiment, we expressed relative growth (g, milligrams per milligram per day) and consumption rates (i, milligrams per milligram per day) as proportions of mean body mass during an experiment using equations with a similar form:

$$(2) \qquad g = \frac{\text{mass}_{\text{fin}} - \text{mass}_{\text{init}}}{((\text{mass}_{\text{fin}} + \text{mass}_{\text{init}})/2)\cdot 7 \text{ days}}$$

and

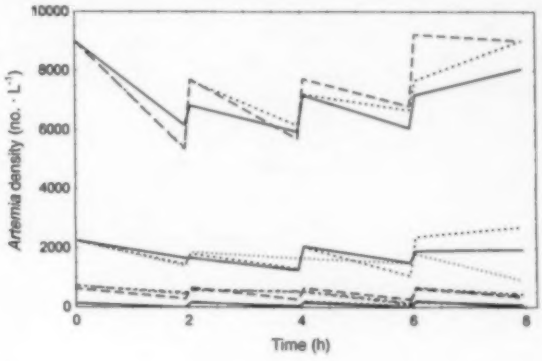
$$(3) \qquad i = \frac{\sum_j^7 (\text{mass eaten on day } j)/\text{NF}_j}{((\text{mass}_{\text{fin}} + \text{mass}_{\text{init}})/2)\cdot 7 \text{ days}}$$

where mass_{fin} and $\text{mass}_{\text{init}}$ were the final and initial masses of the fish in each treatment and NF_j equaled the number of fish in the tank on day j.

We fit type II functional response curves (Holling 1959) to describe the relationship between number of *Artemia* spp. eaten per fish per day (y) and actual mean prey density (x) and between ingestion (milligrams per milligram per day) and *Artemia* density (Proc Nlin, SAS Institute 1989). The form of the equation was

$$(4) \qquad y = \frac{aT_t x}{1 + aT_h x}$$

Fig. 1. *Artemia* density fluctuations over time for a typical day of feeding (20-mm alewife experiment, day 4). Tanks were replenished every 2 h. Each line represents a replicate of one of the four prey densities during an actual day of feeding.



where a and T_h were parameters and T_t was a constant (the number of seconds during feeding (8 h)). Parameter a in eq. 4 represented the instantaneous rate of discovery (attacks per second), and T_h was the handling time per prey (seconds).

The relationship between growth (y, milligrams per day or milligrams per milligram per day) and the independent variables prey density or ingestion (x) was expressed as

$$(5) \qquad y = a(1 - \exp^{-bx}) - c$$

where a, b, and c were parameters. $a - c$ estimated maximum growth rates and c equaled mass loss during starvation when eq. 5 described growth as a function of ingestion. When the relationship between growth and ingestion did not reach an asymptote, we estimated maximum growth from growth at the largest prey density. We calculated maintenance rations (milligrams per milligram per day) by setting y in eq. 5 to zero and solving for x.

To test for the effect of species across the range of experimental prey densities, we fit full and reduced nonlinear models (eq. 4 or 5, Ratkowski 1983) to the relationships described above. In this analysis, we compared the error terms of a reduced model, where a regression was fit to data from all three species together, with recombined full models, where the regression was fit to each species separately and the error terms were summed. The summed error terms from the full models gave an estimate of the total error within species, and the reduced model error term was an estimate of the error among species. The difference between these two error terms yielded an estimate of the gain in precision from fitting species separately and was divided by the reduced minus the summed, full-model degrees of freedom to give the mean square error (MSE) for the difference. An F test, comparing the difference MSE with the reduced MSE, indicated whether we could reject the null hypothesis of no species differences.

Gross growth efficiency (GGE) equals growth divided by ingestion (g/i) and is useful for comparisons because it standardizes growth per unit ingestion. To compare GGE among species, we calculated mean and maximum GGEs for both experiments. We did not report GGE statistics because they were identical to those of the growth versus relative ingestion (g–i) curves. In the 10-mm experiment, maximum GGE equaled the slope of the g–i curve where a straight line with origin zero was tangent to the curve (calculated as the value of i where the derivative of the GGE versus i curve = 0) . Because maximum GGE was not at an intermediate relative ingestion value for the 20-mm fish, we reported maximum GGE for

© 1997 NRC Canada

Table 1. Initial conditions (mean±SD) for each species and experiment.

	Hatch length (mm)	Nominal length	*n*	Actual length (mm)	Initial mass (mg dry)	Age (days)
Alewife	3.9±0.45	10	13	10.6±1.2	0.33±0.10	13
		20	24	20.9±0.72	5.69±0.91	28
Bloater	9.26±0.28	10	21	10.4±0.32	0.90±0.11	3
		20	24	19.9±0.88	4.19±0.77	28
Yellow perch	5.93±0.21	10	24	10.0±0.72	0.87±0.27	18
		20	24	18.6±0.93	8.03±1.47	41

Table 2. Nominal and actual prey densities (no.·L^{-1}).

Nominal treatment	Alewife		Bloater		Yellow perch	
	10 mm	20 mm	10 mm	20 mm	10 mm	20 mm
10	10.8±0.4	24.3±0.1	9.6±0.4	20.5±0.5	13.7±0.6	19.0±0.3
50	53.2±1.9	81.0±0.0	54.3±3.2	77.2±4.3	45.9±0.7	77.5±1.6
250	253.2±8.9	233.3±8.9	262.6±5.4	248.9±3.7	232.2±3.8	222.0±5.5
1000	1018.1±28.9	1028.7±17.0	1044.4±17.1	995.7±31.0	992.3±16.2	964.1±15.2

Note: Values are means ± SD from the 7-day average prey density of the three replicates. Means for each tank were determined daily from the eight density estimates as shown in Fig. 1.

the 20-mm experiments from the feeding level treatment with the highest average GGE.

To compare short-term (15–30 min, Miller et al. 1992) with longer term (8 h, this experiment) functional response predictions, we calculated the number of *Artemia* spp. that a fish would eat in 8 h from the short-term estimate and compared this with the actual number eaten in 8 h. The actual 8-h value was from the 1st day of each trial in the experiment described here, when the sizes in the two experiments were the most similar. Miller et al. (1992) conducted 15- and 30-min functional response experiments with the same species and sizes, and in the same laboratory as reported here. In addition to the duration of the trials, the consumption rate estimation methods also differed between Miller et al. (1993) and the results reported here. We measured depletion rates of the prey as the fish fed, while Miller et al. (1993) examined gut contents. Aside from the methodological differences, the major difference between the two studies is that fish feeding for 8 h must also digest and process the prey, while fish feeding for 15 or 30 min likely did not have enough time to process what they ate. This analysis allowed us to assess the applicability of short-term feeding estimates to a full day of feeding.

Finally, we addressed whether it was possible to detect species differences in survival by incorporating the parameter errors from the laboratory-derived ingestion and growth functions into a version of an individual-based growth and survival model (Letcher et al. 1996). With the model, we tracked the feeding, growth, and survival of individual fish that could die from either predation or starvation. During runs, fish growth was calculated from ingestion rates estimated by the functional responses at two prey densities (10 and 250·L^{-1}). Model runs lasted 7 days and started with 100 fish. Prey were encountered randomly by individual fish (from a Poisson distribution) and both predation (60-mm fish predator) and starvation were size dependent (see Letcher et al. 1996 for details).

Parameter estimates for individual fish were drawn from a normal distribution using means and errors from the ingestion and growth functions. Because the parameters were highly correlated, fish were only allowed combinations of parameters that yielded an r^2 greater than 0.9 for each function. Values of r^2 were estimated from equations (second-order quadratics) relating the error sums of squares to each function's parameter values. With this approach, we avoided parameter combinations yielding unrealistic values.

With the model, we tested how species differences in survival varied with fish size (10 versus 20 mm), prey density (10 and 250·L^{-1}), and number of samples (replicate runs, 3–25). By analyzing increasing numbers of runs, we asked how many samples would be required to detect species differences for small and large fish at low and high prey density.

Results

Actual initial lengths in the experiments were ±1 mm of target lengths, except for 20-mm yellow perch, which averaged 18.6 mm (Table 1). Bloater and yellow perch 10-mm initial masses were similar, and about three times heavier than alewife 10-mm mass (Table 1). Ten-millimetre bloater were the youngest (3 days old) and 10-mm alewife and yellow perch were older (13 and 18 days old, respectively). Twenty-millimetre yellow perch were heaviest and alewife and bloater were 71 and 52% as heavy, respectively. Twenty-millimetre alewife and bloater were both younger (28 days old) than 20-mm yellow perch (41 days old).

During trials, prey densities fluctuated as the prey were eaten and replenished; for most trials the actual daily average prey densities were maintained within 25% of nominal (Fig. 1; Table 2). Food was always available for fish in the 250 and 1000·L^{-1} treatments but was depleted occasionally between feedings in the 50·L^{-1} treatment, and usually in the 10·L^{-1} treatment (Fig. 1), although never for >1 h. Owing to attempts to maintain prey densities, calculated actual densities (Table 2) were about twice the nominal value for the 20-mm fish in the 10 and 50·L^{-1} treatments. Analyses below used actual prey densities.

The functional responses for each experiment were described well by the type II functional response equation

© 1997 NRC Canada

Table 3. Functional response parameters (a, T_h, see eq. 4, Fig. 2) with associated r^2, and maximum consumption rate from this experiment.

	Length (mm)	r^2	$a \pm$ SE ($s^{-1} \times 10^{-4}$)	$T_h \pm$ SE (s)	C_{max} ($mg \cdot mg^{-1} \cdot day^{-1}$)	No. eaten ($(8\ h)^{-1}$) at 1000 prey/L	No. eaten ($(8\ h)^{-1}$) predicted from 15-min experiment
Alewife	10	0.94	1.05±0.35	33.74±5.69	1.89	706.6	285.9
	20	0.99	5.96±0.92	24.09±1.24	0.204	1160.4	7454.3
Bloater	10	0.97	0.94±0.21	23.34±3.46	1.19	380.4	233.6
	20	0.98	7.31±1.32	17.82±1.07	0.434	1882.2	8800.3
Yellow perch	10	0.98	2.53±0.56	54.66±3.81	0.558	535.4	333.7
	20	0.99	8.52±1.24	10.93±0.62	0.296	1854.2	8738.1

Note: The last two columns contain a comparison of actual 8-h consumption rates at 1000 prey/L on day 1 of the experiments presented in this paper and predicted 8-h consumption rates from 15-min functional response experiments (Miller et al. 1992).

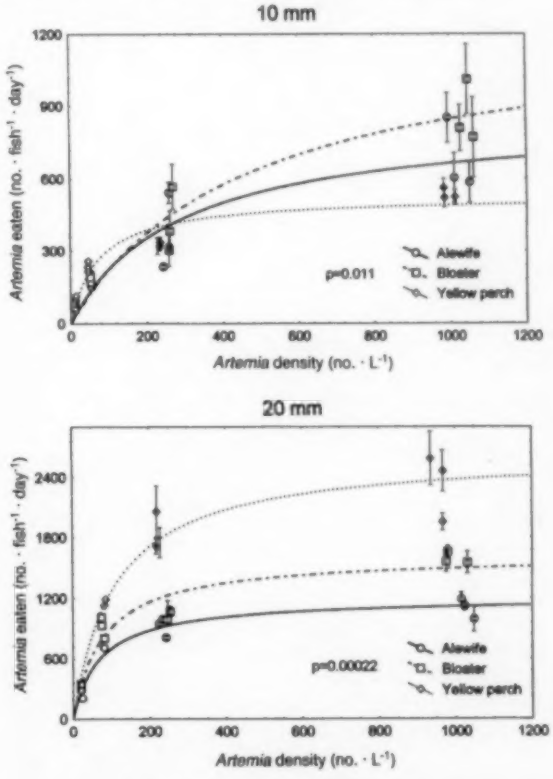
(eq. 2; Table 3, r^2 range of 0.94–0.99). The attack constant, a, was consistently higher for the larger fish and ranged from about 1 to $2.5 \cdot s^{-1}$ for 10-mm fish and from about 6 to $8.5 \cdot s^{-1}$ for 20-mm fish (Table 3). Within a species, handling times (T_h) were always shorter for the larger fish. Ten-millimetre fish required from 23 to 55 s to handle an *Artemia* nauplius, while the larger 20-mm fish took 11–24 s/prey (Table 3). While the number of *Artemia* spp. eaten per 8 h was similar among the species at low prey densities, there were significant differences among species across all prey densities for both 10-mm fish ($p = 0.011$, Fig. 2) and 20-mm fish ($p = 0.0002$, Fig. 2). Twenty-millimetre yellow perch ate the most *Artemia* spp. at all prey densities and alewife ate the least.

When consumption rates were calculated per unit fish mass (relative ingestion), the differences among species were exaggerated ($p = 0.00045$, 10 mm; $p = 0.00017$, 20 mm) relative to calculations using numbers eaten, indicating that functional response differences among species could not be explained by differences in fish masses. Maximum consumption ranged from 20 to 189% body weight·day^{-1} (Table 3).

Comparison of predicted 8-h feeding rates from short-term functional response estimates (15–30 min, Miller et al. 1992) with longer term (8 h) consumption rates revealed that the short-term functional response experiments underestimated by about 33–75% the number actually eaten in 8 h by the 10-mm fish, but overestimated by four- to six-fold the number eaten by 20-mm fish (Table 3). This suggests that smaller 10-mm fish can maintain and exceed short-term feeding rate estimates during a full 8 h of feeding, while larger 20-mm fish cannot.

There were significant absolute growth rate differences among species for both the 10- and 20-mm fish ($p < 0.0001$, Fig. 3). Growth increased rapidly at low prey densities and reached a plateau at the $250 \cdot L^{-1}$ treatment in most cases (Fig. 3). For both lengths, yellow perch grew the fastest at most prey densities. The growth rates of alewife and bloater were similar at 10 mm, while 20-mm alewife grew faster than 20-mm bloater. The magnitude of the species effect was similar ($p < 0.0001$) when the effect of mass was removed by expressing growth rates as a proportion of body mass (Fig. 4). Bloaters consistently grew more slowly at high prey densities than both alewife and yellow perch.

Fig. 2. Functional responses for the six experiments. Each point represents the mean value (±SE) of an individual tank. The p values are from the full-reduced nonlinear test of the null hypothesis of no species differences in functional responses. Parameter values are in the Appendix and error bars were omitted for lower prey densities for clarity.



Plotting relative growth versus relative ingestion removes the effect of prey density (and fish mass) and shows the relationship between food consumed and growth. There were significant differences among species, although differences

© 1997 NRC Canada

Fig. 3. Absolute growth rates (means ± SE) as a function of prey density for the six experiments. The p values are from the full-reduced nonlinear test of the null hypothesis of no species differences in growth rates.

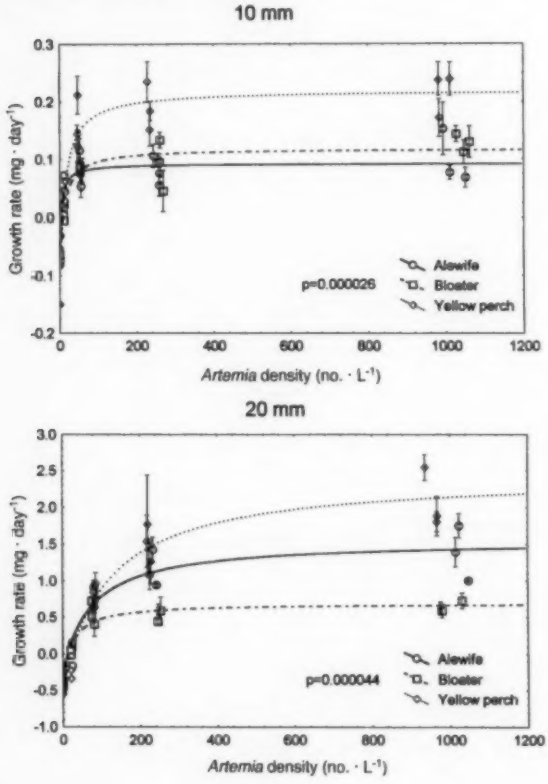
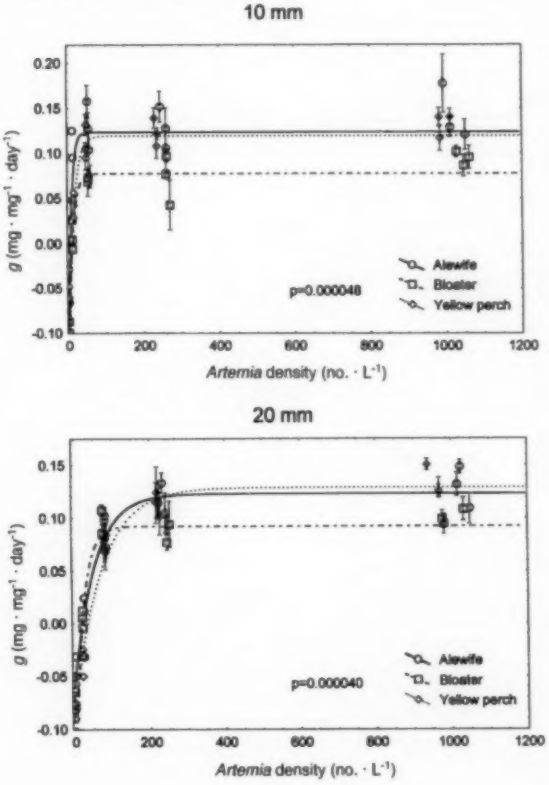


Fig. 4. Relative growth rates (means ± SE) as a function of prey density for the six experiments. The p values are from the full-reduced nonlinear test of the null hypothesis of no species differences in growth rates.



were weaker for 10-mm fish (Fig. 5, $p = 0.021$) than for 20-mm fish (Fig. 5, $p = 0.0006$). Ten-millilitre bloater grew the slowest per unit ingestion, especially at higher ingestion rates, while alewife and yellow perch grew at similar rates. Twenty-millilitre alewife grew from approximately 0.05 to 0.07·day^{-1} faster than did bloater or yellow perch.

The g–i curves for the 10-mm fish flattened at high ingestion rates, which is typical for larval fish (Kiorboe et al. 1987; Letcher and Bengtson 1993*a*), but the curves of the 20-mm fish were linear (alewife and yellow perch) or slightly decelerating (bloater) over the range of ingestion rates observed (Fig. 5). It is unlikely that fish were limited by prey density, which would have limited ingestion rates and the possibility of maximum growth rate, because food was constantly available during the 8-h feeding periods at the two higher feeding levels (250 and 1000·L^{-1}). The differences in curve shape may be related to developmental stage; both the 20-mm alewife and yellow perch were near metamorphosis (Auer 1982; B.H. Letcher, personal observation), the period of development when gut morphology changes rapidly.

Mean and maximum GGEs of the 20-mm fish were about twofold greater than those of the 10-mm fish (Table 4). Alewife consistently had the highest mean GGEs (average = 0.25), bloater's was lower (average = 0.15), and the average maximum GGE of yellow perch was intermediate (average = 0.19) (Table 4). Overall, alewife converted food to body mass most efficiently, while bloater were the least efficient.

With our model simulations, we tested the effects of fish size, prey density, and number of samples on our ability to detect differences in survival among species. There were significant differences among species ($p < 0.001$, one-way ANOVA) for both prey densities (10 and 250·L^{-1}) and all number of samples (3–25) for 20-mm fish, suggesting that species differences in ingestion and growth rates are very likely to translate into differences in survival for 20-mm fish. In contrast, there was an interaction between prey density and number of samples for the 10-mm fish; significant differences ($p < 0.05$) in survival among species occurred with fewer samples (<3, p value = 0.038 – 0.002(number of samples), $p < 0.002$) for fish at 10 prey/L, while 23 samples were required for fish at 250 prey/L (p value = 0.605 – 0.025(number of samples), $p < 0.0001$). Overall, model results suggest that we are likely to find species differences in survival for 20-mm fish and for 10-mm fish at low prey densities, but that species differences will be difficult to detect for 10-mm fish at higher prey densities.

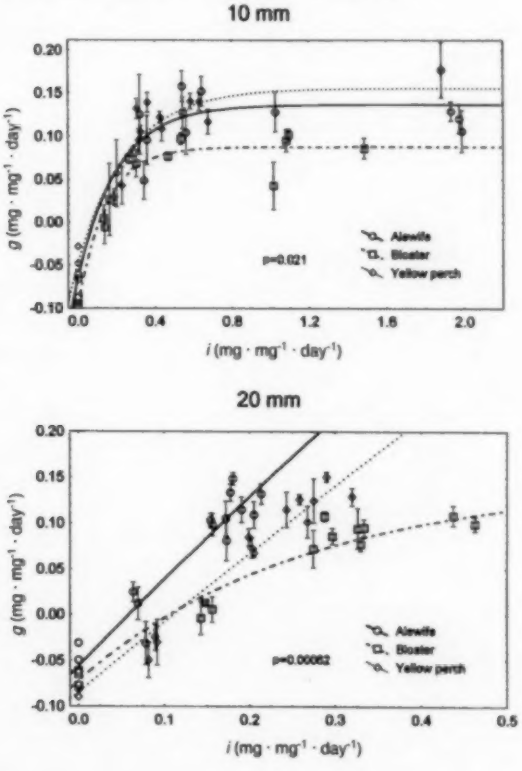
© 1997 NRC Canada

Table 4. Growth and consumption rate data for the six experiments.

	Length (mm)	Absolute starving mass loss ($mg \cdot day^{-1}$)	Relative starving mass loss ($mg \cdot mg^{-1} \cdot day^{-1}$)	Maintenance ration ($mg \cdot mg^{-1} \cdot day^{-1}$)	Maximum growth ($mg \cdot mg^{-1} \cdot day^{-1}$)	GGE (mean ± SD)	Maximum GGE	i at maximum GGE ($mg \cdot mg^{-1} \cdot day^{-1}$)
Alewife	10	0.030	0.0643	0.0346	0.137	0.146±0.104	0.309	0.237
	20	0.252	0.0559	0.0254	Undefined	0.452±0.326	Undefined	Undefined
Bloater	10	0.070	0.0692	0.0303	0.115	0.127±0.100	0.215	0.259
	20	0.422	0.0717	0.0461	0.148	0.167±0.115	0.254	0.315
Yellow perch	10	0.095	0.0482	0.0301	0.156	0.135±0.148	0.315	0.237
	20	0.480	0.0842	0.0479	Undefined	0.251±0.289	Undefined	Undefined

Note: Starving mass loss ($mg \cdot day^{-1}$) is the absolute mass loss per day and relative starving mass loss ($mg \cdot mg^{-1} \cdot day^{-1}$) equals c in eq. 5. Maximum growth was estimated as $a - c$ from eq. 5 (undefined when the function did not asymptote over the range of data), and maintenance ration is the proportion of body mass required to maintain constant body mass. Mean GGE was calculated over all treatments; maximum GGE represents the greatest predicted GGE for each experiment and ingestion (i) at maximum GGE is the relative daily consumption rate required to give maximum GGE.

Fig. 5. Relative growth rates (means ± SE) as a function of relative ingestion rates for the six experiments. The p values are from the full-reduced nonlinear test of the null hypothesis of no species differences in the $g - i$ relationship.



Discussion

We investigated the effects of species and size on ingestion and growth rates for three species of Lake Michigan fish that hatch over a broad range of sizes (3.5–9.5 mm). Our null hypothesis was that species differences would have no effect on ingestion and growth rates for fish of similar sizes. Growth and ingestion rates were significantly different for both 10- and 20-mm fish, although differences among species were stronger for the larger fish. Incorporating the growth and ingestion relationships and their parameter errors into a population simulation model demonstrated that we are likely to detect species differences in survival for 20-mm fish, but that detecting differences for 10-mm fish at high prey density would require an unrealistically high number of samples (>23). Thus, while there may be size-based differences among small fish that are generalizable across taxa (Miller et al. 1988; Houde 1989; Houde and Zastrow 1993), differences among species will contribute to a large part of the variability in survival among species, especially for fish >10 mm (Houde and Schekter 1980, 1981).

Small differences in growth among populations (Conover 1990), cohorts (Leak and Houde 1987; Kneib 1993), or individuals (Rosenberg and Haugen 1982; Rice et al. 1993; Rose and Cowan 1993) within a species are more likely to obscure species differences when fish are <10 mm than when they are larger. As fish approach metamorphosis, species differences become more pronounced (Fig. 5). Investigating the effects of variability in growth within species for small fish (<10 mm) and both within and among species for larger fish (>20 mm) may lead to insights about processes that control survival during early life history. In general, differences among freshwater larvae, which are larger at hatch, may be more pronounced than differences among marine larvae (Houde 1994).

Estimates of species effects derived under identical conditions in the laboratory may be conservative relative to differences that could exist in the field. Species differences in nature may be magnified because the fish in this study could not choose habitat or prey types. The differences among species observed in this study represent alternative physiologies and bioenergetics, and possibly not the full range of among-species variability because behavioral and habitat choices were constrained.

Alternatively, species performance (consumption and growth rates) could converge in nature if the full range of behavioral and habitat choices were available. For instance,

© 1997 NRC Canada

different thermal optima among species could result in varying responses. If the thermal optimum for a species is lower than the temperature used here (22°C), the performance of that species may be compromised by the relatively high temperature. This may have been the case for the bloater, which hatch at lower temperatures (2–4°C, Wells 1966), although bloater do move into warmer surface waters within 1–2 weeks of hatching. Optimal temperature for food consumption by adult bloater is at least 16.8°C (maximum temperature in Binkowski and Rudstam 1994). Alewife and yellow perch, which occupy the littoral zone during early life and therefore typically may encounter slightly higher temperatures and do appear to have higher optimal consumption temperatures (24 and 29°C, respectively (Post 1990; Stewart and Binkowski 1986)), grew at similar relative rates as a function of prey density, and both species grew faster than bloater (Fig. 4). The extent to which evaluating species effects at a single temperature affected our conclusions is unknown. In contrast with growth and consumption as functions of prey density, the relationship between relative growth and ingestion does not appear to be affected by temperature (Checkley 1984; Houde 1989; Houde and Zastrow 1993; Letcher and Bengtson 1993*a*). Differences in thermal optima among species, therefore, may be relatively unimportant in regulating species differences in growth efficiency.

Miller et al. (1992) investigated the size dependence of parameters of the type II functional response curve over a wide range of young-of-year lengths for the same three species used here. They found no significant effect of species on parameter estimates for attack constant and handling time for fish from 10 to 40 mm, but there was a significant effect of size. This contrasts with the results presented here where, within sizes, we found a significant effect of species on functional responses. The different conclusion between the two studies probably reflects differences in statistical design and level of analysis. We were interested in the level of complexity required to predict growth and consumption rates of fish of different sizes. For predictive ability, we found that we needed to include species in our description of consumption rate. Miller et al. (1992) asked questions on a much broader scale (across sizes, not within) and over a wider range of sizes, with the goal of evaluating how much variability in consumption rate parameters could be explained by size and species. Clearly, results depend on the questions asked and on the level of analysis.

The shape of the curves that relate growth to ingestion (g–i curves) appears to depend on developmental stage. Larval fish had decelerating g–i curves, but fish close to or beyond metamorphosis had linear g–i curves (Fig. 5). Alewife and yellow perch both metamorphose around 19–20 mm (Auer 1982), while bloater metamorphose between 25 and 30 mm (Auer 1982). The g–i curve for 25-mm yellow perch was also linear (B.H. Letcher, unpublished results). Further, the temperature-corrected summary relationships for many species of larvae reported by Checkley (1984) and Letcher and Bengtson (1993*a*) also showed decelerating relationships. The decelerating g–i curves of the larval fish suggest that they maximize consumption rate and not efficiency of growth (Checkley 1984). Despite decreasing increments in growth per unit increase in ingestion, larvae maintained rapid ingestion rates at high prey densities. Larvae may have evolved to maximize growth rate over efficiency because of the strongly size-dependent nature of larval mortality (Miller et al. 1988). The larger, metamorphosed fish (20-mm alewife and yellow perch) seemed to consume prey at a rate that maximized conversion efficiency (linear g–i curves, Fig. 5). The conversion efficiencies of juvenile fish may be higher for at least two reasons; ingestion rates of metamorphosed fish may be limited by the development of differentiated guts, and increased digestive efficiency of the older fish (Buckley and Dillmann 1982; Govoni et al. 1982; Klumpp and von Westernhagen 1986) may allow better processing of the meal.

Many functional response estimates in the literature are from short-term feeding experiments (e.g., Miller et al. 1992). This is often the case because trials must be short enough to avoid prey depletion and gut evacuation when feeding rates are estimated from gut contents. These short-term functional response estimates frequently are used to estimate feeding over an entire day of feeding. However, short-term estimates may not be good predictors of full-day ingestion rates if feeding rate is not constant during a whole day of feeding. Comparison between short-term (15–30 min, Miller et al. 1992) feeding rate estimates based on gut contents and the longer term (8 h) prey depletion estimates presented in this paper revealed that the short-term estimates underestimated full-day feeding rates by 33–75% for 10-mm larvae, but they overestimated full-day feeding rates by four to six times for 20-mm larvae (Table 3). These results are probably related to digestion differences between 10- and 20-mm fish. Because the digestive systems of the 20-mm fish had begun to develop into stomachs (alewife and yellow perch had metamorphosed), the larger fish were likely eating meals instead of feeding continuously. Short-term 15- to 30-min trials may measure the rate at which juveniles fill their guts without accounting for the time required for digestive pauses. This would result in elevated short-term ingestion rate estimates relative to the longer term estimates.

The opposite pattern was observed with the 10-mm fish; the short-term estimates underestimated full-day estimates. This suggests that 10-mm larvae were capable of maintaining and even exceeding the short-term feeding rate estimates during a full 8 h of feeding. High, sustained ingestion rates of larvae are consistent with their gut morphology (tube, no stomach) and a "conveyor-belt approach" to digestion. High ingestion rates of 10-mm larvae are also consistent with the notion that larvae maximize feeding rate (see above). In support of this view, Letcher and Bengtson (1993*b*) found that 7-day-old larval inland silverside (*Menidia beryllina*) consumed prey at a constant rate during 12 h of feeding in the laboratory. These results indicate that adequately predicting full-day feeding rates from short-term functional response experiments may depend on the developmental stage of the fish.

Our results indicate that we will be unlikely to predict growth and consumption rates of the three species we studied on the basis of size alone; species-specific relationships appear to be required. Although differences in growth and consumption rates among species were less pronounced for the 10-mm fish, they were still statistically significant. Simulation model results using the data from this study indicated that species differences in survival may be readily detectable for 20-mm fish, but were not always so for 10-mm fish. Gen-

© 1997 NRC Canada

erally, it appears that we will not be able to use common, size-dependent relationships to predict growth, consumption, or survival rates for small fish (<20 mm), although differences in growth and survival among 10-mm fishes will be more difficult to detect than differences among 20-mm fishes.

Acknowledgments

We thank Thomas Miller and two anonymous reviewers for constructive comments on the manuscript. This work would not have been possible without the technical assistance of Marsha Jones and Brian Nordskog. Julia Auckland and Elizabeth Priddy tirelessly counted the samples of uneaten brine shrimp nauplii. We thank the staff of The Center for Great Lakes Studies, in particular Chris Ney, Jason Kroll, Dave Meuller, and Steve Yeo, who collected and fertilized the zygotes, often under difficult circumstances, and who raised the fish and kept things running smoothly. This work was funded by the University of Wisconsin Sea Grant Institute under grants from the National Sea Grant College Program, National Oceanic and Atmospheric Administration, U.S. Department of Commerce, and from the State of Wisconsin, Federal grant No. NA90AA-D-SG469, project No. NA-84AA-D-00065, and by an Electric Power Research Institute fellowship to B.H.L.

References

Anderson, J.T. 1988. A review of size dependent survival during pre-recruit stages of fishes in relation to recruitment. J. Northwest Atl. Fish. Soc. **8**: 55–66.

Auer, N.A. 1982. Identification of larval fishes of the Great Lakes basin with emphasis on the Lake Michigan drainage. Special publication 82-3 edition. Great Lakes Fishery Commission, Ann Arbor, Mich.

Beyer, J.E. 1989. Recruitment stability and survival: simple size-specific theory with examples from the early life dynamics of marine fish. Dana, **7**: 45–147.

Blaxter, J.H.S. 1986. Development of sense organs and behaviour of teleost larvae with special reference to feeding and predator avoidance. Trans. Am. Fish. Soc. **115**: 98–114.

Binkowski, F.P., and Rudstam, L.G. 1994. Maximum daily ration of Great Lakes bloater. Trans. Am. Fish. Soc. **123**: 335–342.

Buckley, L.J., and Dillmann, D.W. 1982. Nitrogen utilization by larval summer flounder *Paralichthys dentatus* (Linnaeus). J. Exp. Mar. Biol. Ecol. **59**: 243–256.

Checkley, D.M. 1984. Relation of growth to ingestion for larvae of Atlantic herring *Clupea harengus* and other fish. Mar. Ecol. Prog. Ser. **18**: 215–224.

Conover, D.O. 1990. The relation between capacity for growth and length of growing season: evidence for and implications of countergradient variation. Trans. Am. Fish. Soc. **119**: 416–430.

Govoni, J.J., Peters, D.S., and Merriner, J.V. 1982. Carbon assimilation during larval development of the marine teleost *Leiostomus xanthurus* Lacepede. J. Exp. Mar. Biol. Ecol. **64**: 287–299.

Holling, C.S. 1959. The components of predation as revealed by a study of small mammal predation of the European sawfly. Can. Entomol. **91**: 293–332.

Houde, E.D. 1987. Fish early life dynamics and recruitment variability. Am. Fish. Soc. Symp. **2**: 17–29.

Houde, E.D. 1989. Comparative growth, mortality, and energetics of marine fish larvae: temperature and implied latitudinal effects. Fish. Bull. **87**: 471–495.

Houde, E.D. 1994. Differences between marine and freshwater fish larvae: implications for recruitment. ICES J. Mar. Sci. **51**: 91–97.

Houde, E.D., and Schekter, R.C. 1980. Feeding by marine fish larvae: developmental and functional responses. Environ. Biol. Fishes, **5**: 315–334.

Houde, E.D., and Schekter, R.C. 1981. Growth rates, rations and cohort consumption of marine fish larvae in relation to prey concentrations. Rapp. P.-V. Reun. Cons. Int. Explor. Mer, **178**: 441–453.

Houde, E.D., and Zastrow, C.E. 1993. Ecosystem- and taxon-specific dynamic and energetics properties of larval fish assemblages. Bull. Mar. Sci. **53**: 290–335.

Kiorboe, T., Munk, P., and Richardson, K. 1987. Respiration and growth of larval herring *Clupea harengus*: relation between specific dynamic action and growth efficiency. Mar. Ecol. Prog. Ser. **40**: 1–10.

Klumpp, D.W., and von Westernhagen, H. 1986. Nitrogen balance in marine fish larvae: influence of developmental stage and prey density. Mar. Biol. (Berlin), **93**: 189–199.

Kneib, R.T. 1993. Growth and mortality in successive cohorts of fish larvae within an estuarine nursery. Mar. Ecol. Prog. Ser. **94**: 115–127.

Leak, J.C., and Houde, E.D. 1987. Cohort growth and survival of bay anchovy *Anchoa mitchilli* larvae in Biscayne Bay, Florida. Mar. Ecol. Prog. Ser. **37**: 109-122.

Letcher, B.H. 1994. Causes and consequences of growth variability in young fishes. Ph.D. dissertation, North Carolina State University, Raleigh, N.C.

Letcher, B.H., and Bengtson, D.A. 1993a. Effects of food density and temperature on feeding and growth of young inland silversides (*Menidia beryllina*). J. Fish Biol. **43**: 671–686.

Letcher, B.H., and Bengtson, D.A. 1993b. Effects of food density on growth and patterns of food consumption by larval silverside fish, *Menidia beryllina*; a laboratory investigation with image analysis. J. Exp. Mar. Biol. Ecol. **167**: 197–213.

Letcher, B.H., Rice, J.A., Crowder, L.B., and Rose, K.A. 1996. Variability in survival of larval fishes: disentangling components with a generalized individual-based model. Can. J. Fish. Aquat. Sci. **53**: 787–801.

Miller, T.J., Crowder, L.B., Rice, J.A., and Marschall, E.A. 1988. Larval size and recruitment mechanisms in fishes: toward a conceptual framework. Can. J. Fish. Aquat. Sci. **45**: 1657–1670.

Miller, T.J., Crowder, L.B., Rice, J.A., and Binkowski, F.P. 1992. Body size and the ontogeny of the functional response in fishes. Can. J. Fish. Aquat. Sci. **49**: 805–812.

Miller, T.J., Crowder, L.B., and Rice, J.A. 1993. Ontogenetic changes in behavioural and histological measures of visual acuity in three species of fish. Environ. Biol. Fishes, **37**: 1–8.

Post, J.R. 1990. Metabolic allometry of larval and juvenile yellow perch (*Perca flavescens*): in situ estimates and bioenergetic models. Can. J. Fish. Aquat. Sci. **47**: 554–560.

Ratkowski, D.A. 1983. Nonlinear regression modeling. Marcel Dekker, New York.

Rice, J.A., Miller, T.J., Rose, K.A., Crowder, L.B., Marschall, E.A., Trebitz, A.S., and DeAngelis, D.L. 1993. Growth rate variation and larval survival: inferences from an individual-based size-dependent predation model. Can. J. Fish. Aquat. Sci. **50**: 133–142.

Rose, K.A., and Cowan, J.H., Jr. 1993. Individual-based model of young-of-the-year striped bass population dynamics. I. Model description and baseline simulations. Trans. Am. Fish. Soc. **122**: 415–438.

Rosenberg, A.A., and Haugen, A.S. 1982. Individual growth and size-selective mortality of larval turbot (*Scophthalmus maximus*) reared in enclosures. Mar. Biol. (Berlin), **72**: 73–77.

SAS Institute. 1989. SAS user's guide, version 6. SAS Institute, Cary, N.C.

© 1997 NRC Canada

Sissenwine, M.P. 1984. Why do fish populations vary? *In* Exploitation of marine communities. *Edited by* R. May. Springer-Verlag, Berlin. pp. 59–94.

Stewart, D.J., and Binkowski, F.P. 1986. Dynamics of consumption and food conversion by Lake Michigan alewives: an energetics-modeling synthesis. Trans. Am. Fish. Soc. **115**: 643–661.

Wells, L. 1966. Seasonal and depth distribution of larval bloater (*Coregonus hoyi*) in southeastern Lake Michigan. Trans. Am. Fish. Soc. **95**: 388–396.

Appendix

Parameters (±SE) for the equations describing feeding and growth relationships (Figs. 2–5).

Alewife			
	a	*b*	*c*
10-mm experiment			
Ingestion ($mg \cdot mg^{-1} \cdot day^{-1}$) vs. prey density	$4.34 \times 10^{-7} \pm 2.0 \times 10^{-7}$	12 933.6 ± 1 922.2	0
Growth ($mg \cdot day^{-1}$) vs. prey density	0.116 ± 0.009 4	0.097 5 ± 0.039	0.030 ± 0.000 42
Growth ($mg \cdot mg^{-1} \cdot day^{-1}$) vs prey density	0.246 ± 0.006 84	0.135 ± 0.020 9	0.122 ± 0.005 40
Growth ($mg \cdot mg^{-1} \cdot day^{-1}$) vs. ingestion ($mg \cdot mg^{-1} \cdot day^{-1}$)	0.201 ± 0.019 2	4.842 ± 1.389	0.064 0 ± 0.015 6
20-mm experiment			
Ingestion ($mg \cdot mg^{-1} \cdot day^{-1}$) vs. prey density	$1.98 \times 10^{-7} \pm 0.1 \times 10^{-7}$	136 276.1 ± 5 588.49	0
Growth ($mg \cdot day^{-1}$) vs. prey density	1.597 ± 0.106	0.011 3 ± 0.002 43	0.252 ± 0.057 7
Growth ($mg \cdot mg^{-1} \cdot day^{-1}$) vs. prey density	0.178 ± 0.008 25	0.017 8 ± 0.003 03	0.055 1 ± 0.014 6
Growth ($mg \cdot mg^{-1} \cdot day^{-1}$) vs. ingestion ($mg \cdot mg^{-1} \cdot day^{-1}$)	2.070 ± 15.500	0.468 ± 3.673	0.055 9 ± 0.014 2

Bloater			
	a	*b*	*c*
10-mm experiment			
Ingestion ($mg \cdot mg^{-1} \cdot day^{-1}$) vs. prey density	$1.76 \times 10^{-7} \pm 1.0 \times 10^{-7}$	18 419.4 ± 2 989.5	0
Growth ($mg \cdot day^{-1}$) vs. prey density	0.176 ± 0.008 8	0.056 8 ± 0.014 4	0.070 ± 0.003 74
Growth ($mg \cdot mg^{-1} \cdot day^{-1}$) vs prey density	0.182 ± 0.005 5	0.089 6 ± 0.013 4	0.104 ± 0.006 81
Growth ($mg \cdot mg^{-1} \cdot day^{-1}$) vs. ingestion ($mg \cdot mg^{-1} \cdot day^{-1}$)	0.184 ± 0.012 2	6.767 ± 1.091	0.096 2 ± 0.009 99
20-mm experiment			
Ingestion ($mg \cdot mg^{-1} \cdot day^{-1}$) vs. prey density	$3.18 \times 10^{-7} \pm 1.0 \times 10^{-7}$	63 120.6 ± 3 165.7	0
Growth ($mg \cdot day^{-1}$) vs. prey density	1.030 ± 0.036 5	0.030 1 ± 0.004 58	0.422 ± 0.026 6
Growth ($mg \cdot mg^{-1} \cdot day^{-1}$) vs. prey density	0.220 ± 0.004 09	0.044 4 ± 0.003 91	0.128 ± 0.008 60
Growth ($mg \cdot mg^{-1} \cdot day^{-1}$) vs. ingestion ($mg \cdot mg^{-1} \cdot day^{-1}$)	0.220 ± 0.031 1	3.711 ± 1.113	0.071 7 ± 0.009 46

Yellow perch			
	a	*b*	*c*
10-mm experiment			
Ingestion ($mg \cdot mg^{-1} \cdot day^{-1}$) vs. prey density	$5.40 \times 10^{-7} \pm 1.0 \times 10^{-7}$	49 750.6 ± 3 505.9	0
Growth ($mg \cdot day^{-1}$) vs. prey density	0.297 ± 0.012 0	0.044 6 ± 0.007 3	0.094 8 ± 0.028 8
Growth ($mg \cdot mg^{-1} \cdot day^{-1}$) vs prey density	0.257 ± 0.004 63	0.075 6 ± 0.006 51	0.138 ± 0.048 6
Growth ($mg \cdot mg^{-1} \cdot day^{-1}$) vs. ingestion ($mg \cdot mg^{-1} \cdot day^{-1}$)	0.204 ± 0.025 2	3.893 ± 1.118	0.048 2 ± 0.012 4
20-mm experiment			
Ingestion ($mg \cdot mg^{-1} \cdot day^{-1}$) vs. prey density	$2.28 \times 10^{-7} \pm 0.1 \times 10^{-7}$	92 929.2 ± 3 079.6	0
Growth ($mg \cdot day^{-1}$) vs. prey density	2.541 ± 0.122	0.007 56 ± 0.001 05	0.480 05 ± 0.039 0
Growth ($mg \cdot mg^{-1} \cdot day^{-1}$) vs. prey density	0.206 ± 0.006 32	0.013 5 ± 0.001 44	0.769 ± 0.008 03
Growth ($mg \cdot mg^{-1} \cdot day^{-1}$) vs. ingestion ($mg \cdot mg^{-1} \cdot day^{-1}$)	4.402 ± 34.093	0.175 ± 1.389	0.084 2 ± 0.009 75

Note: The form of the feeding equations was $y = \frac{adx}{1 + abx} + c$ where a, b, and c were parameters, x was prey density (no. $\cdot L^{-1}$), y was feeding rate ($mg \cdot mg^{-1} \cdot day^{-1}$), and d equaled $8 \times 60 \times 60$ (the number of seconds available for feeding each day). We estimated the effect of prey density and feeding rate growth using $y = a(1 - \exp^{-bx}) - c$ where a, b, and c were parameters (Proc Nlin, SAS Institute 1989). For each species-specific estimate the number of observations equaled 15. Functional response parameters for the 10- and 20-mm experiments are given in Table 3.

© 1997 NRC Canada

A demographic method with population density compensation for estimating productivity and yield per recruit of the leopard shark (*Triakis semifasciata*)

David W. Au and Susan E. Smith

Abstract: A procedure is shown, using the leopard shark (*Triakis semifasciata*) as an example, for obtaining estimates of a population's intrinsic rate of increase (r) that depends on the level of total mortality borne. The method uses age at female maturity, maximum reproductive age, and average fecundity and incorporates concepts of density dependence from standard fisheries population modelling. An estimate of the leopard shark's r value at the population size producing maximum sustainable yield was thus obtained and then extended to adjust estimates of its yield per recruit for the effects of recruitment reduction under fishing pressure. The results indicate that this shark may be more susceptible to overexploitation than previously thought. This procedure is shown to be capable of returning r values for the leopard and five other species of sharks that compare favorably with published values obtained by the more standard life table method. It should prove useful for assessing the status of populations for which few data exist.

Résumé : On montre une méthode, en prenant pour exemple le requin-tigre (*Triakis semifasciata*), pour obtenir des valeurs estimées du taux d'augmentation intrinsèque d'une population (r) qui dépend du degré de mortalité total éprouvé. La méthode utilise l'âge de maturité de la femelle, l'âge de reproduction maximal et la fertilité moyenne et incorpore des concepts de dépendance à l'égard de la densité tirés de la modélisation standard des populations de poissons. Une estimation de la valeur r du requin-tigre pour la taille de la population produisant le rendement maximal durable a ainsi été obtenue, puis étendue pour ajuster les valeurs estimées de son rendement par recrue pour tenir compte des effets de la réduction du recrutement attribuable à la pêche. Les résultats indiquent que ce requin pourrait être plus vulnérable à la surpêche qu'on le croyait jusqu'ici. On a montré que cette méthode est capable de donner des valeurs r pour le requin-tigre et cinq autres espèces de requins qui se comparent favorablement avec les valeurs publiées obtenues par la méthode plus traditionnelle des tables de survie. Elle devrait s'avérer utile pour évaluer l'état des populations pour lesquelles il existe peu de données.
[Traduit par la Rédaction]

Introduction

Demographic analyses can be useful to gain understanding of the status of exploited populations for which empirical production and mortality-estimating data are difficult to obtain. Sharks are usually good candidates for such studies for the following reasons: (*i*) their populations are often widely and thinly distributed, with catches poorly reported and the extent of exploitation seldom clear; (*ii*) shark vital rates are often difficult to estimate because of aging and sampling difficulties; and (*iii*) shark births may be proportional to, and subsequent recruitment often dependent on, stock size (Holden 1973), thus complicating the assessments of population status. Moreover, the assumptions in demographic analyses of stable-aged distributions and population changes at unvarying rates do resemble real shark dynamics, for most shark populations appear to grow slowly and steadily, without strongly varying strengths of successive year-classes. Recent demographic studies of sharks that estimate their rates of increase using standard methods are by Jones and Geen (1977), Hoenig and Gruber (1990), Hoff (1990), Cailliet (1992), Cailliet et al. (1992), and Cortes (1995).

In the present study, we also use the demographic approach, but unlike the standard demographic procedure, we do not use the typical life history table methodology. Rather, we develop a modified method that incorporates standard fisheries concepts of density-dependent compensation, which alters vital rates to produce increased productivity at intermediate population sizes. The purpose is to provide a systematic method for estimating a population's productivity, even when comprehensive demographic schedules are lacking.

First, we show a straightforward method of estimating an exploited population's intrinsic rate of increase (r) with minimal data input, and second, we show how this estimate can be used to adjust yield per recruit (Y/R) calculations. This is accomplished by incorporating a stock–recruitment relationship to account for the effects of recruitment being related to population size.

Recruitment is of key interest because if a Y/R diagram is viewed as a comparison of productivities of a standing stock at different population sizes resulting from different combinations of fishing mortality and age at first capture, then

Received February 2, 1996. Accepted July 16, 1996.
J13283

D.W. Au[1] and S.E. Smith. Southwest Fisheries Science Center, National Marine Fisheries Service, P.O. Box 271, La Jolla, CA 92038, U.S.A.

[1] Author to whom all correspondence should be addressed.
e-mail: david.au@noaa.gov

© 1997 NRC Canada

adjustments must be made for any changes in level of recruitment. In this vein, Kimura (1988) recently reemphasized the need to account for density-dependent recruitment changes in population assessments and noted that Beverton and Holt (1957) examined this subject at length in their original discussion of Y/R analysis. Introducing a method not described before, we use the concept of density dependence in estimating r.

We use the leopard shark (*Triakis semifasciata*) as an example. This species is chosen because an estimate of its r value and calculation of its Y/R have been published; it is caught mainly by anglers in California waters (Smith and Abramson 1990; Cailliet 1992).

Methods

Estimating intrinsic rate of increase

The classic demographic relationship for a population can be simplified and the concept of sustainable yield invoked to allow solution for the population's intrinsic rate of increase at a particular level of adult mortality.

Lotka's equation simplified

The relationship between age at maturity (α), maximum reproductive age (w), survival to age x (l_x), fecundity at age x (m_x) (in annual newborn females per adult female), and intrinsic rate of increase (r) for a stable-aged population is expressed by Lotka's (1959) equation (more correctly, the Euler–Lotka equation).This fundamental demographic equation may be written (e.g., Stearns 1992) as

$$(1)\quad \sum_{x=\alpha}^{w} l_x e^{-rx} m_x = 1.0$$

(see Table 1 for a list of parameter and equation symbols). The parameter r is usually solved from this relationship, but here we seek a form less dependent on detailed survival and fecundity schedules that are often lacking for sharks.

First, survival to age x, or l_x, is written in terms of survival to maturity at age α (i.e., l_α). Relative to l_α, l_x is $l_\alpha(l_x/l_\alpha) = l_\alpha e^{-M(x-\alpha)}$, for $x \geq \alpha$ and where M is adult instantaneous natural mortality. The latter can be estimated from maximum age under unfished conditions as described by Hoenig (1983). Equation 1 is now written as

$$(2)\quad l_\alpha \sum_{x=\alpha}^{w} e^{-M(x-\alpha)} e^{-rx} m_x = 1.0.$$

Next, average fecundity of all adult females (b) is substituted for m_x. Equation 2 then becomes

$$l_\alpha b \sum_{x=\alpha}^{w} e^{-M(x-\alpha)} e^{-rx} = l_\alpha b e^{M\alpha} \sum_{x=\alpha}^{w} e^{-(M+r)x} = 1.0$$

or

$$l_\alpha b e^{M\alpha} e^{-(M+r)\alpha} [1 + e^{-(M+r)} + \ldots + e^{-(M+r)(w-\alpha)}] = 1.0.$$

Upon substituting the relationship for a sum of a geometric series for the bracketed terms, and then further simplifying, this becomes

$$l_\alpha b e^{-r\alpha} [(1 - e^{-(M+r)(w-\alpha+1)})/(1 - e^{-(M+r)})] = 1.0$$

Table 1. Parameter and symbol definitions.

Symbol	Definition
r	Intrinsic rate of population increase
r_Z	r when adults suffer mortality rate Z
r_{2M}	r when adults suffer mortality rate $Z = 2M$ (r at MSY)
x	Age
α	Age of females at 50% maturity ($x = \alpha$)
w	Maximum reproductive age ($x = w$)
l_x	Survival to age x
l_α	Survival to age at maturity α (preadult or juvenile survival)
$l_{\alpha,Z}$	l_α when adults suffer mortality rate Z
$l_{\alpha,2M}$	l_α when adults suffer mortality rate $Z = 2M$ (l_α at MSY)
m_x	Fecundity (female pups/female) at age x
b	Average fecundity of adult females
M	Instantaneous adult natural mortality rate
F	Instantaneous fishing mortality rate
F-max	The F giving maximum Y/R at any given age at first capture
Z	Instantaneous adult total mortality rate ($Z = M + F$)
S–R	Stock–recruitment relationship
R'	Fractional recruitment level (fraction of unexploited level)
P'	Fractional stock level (fraction of unexploited level)
A	Ricker (1975) shape parameter for Beverton and Holt (1957) S–R relationship
μ_s	Rate of sustainable exploitation for MSY
e^r	Annual rate of population increase
$e^{r_{2M}}$	Annual rate of increase for population giving MSY
$t_{r'}$	Age at first capture
Y/R	Yield per recruit
Y/RA	Recruitment-adjusted yield per recruit

(the denominator not equalling zero), or

$$(3)\quad e^{-(M+r)} + l_\alpha b e^{-r\alpha} [1 - e^{-(M+r)(w-\alpha+1)}] = 1.0.$$

Equation 3 is a simplified form of Lotka's equation from which r can be estimated using the five life history parameters of age at maturity α, maximum reproductive age w, adult instantaneous natural mortality M, average number of female pups per adult female b, and survival to age at maturity l_α. A similar but more complicated equation, in terms of survival to age 1 rather than to age at maturity α, was developed by D.S. Vaughan (described by Hoenig and Gruber 1990).

Mortality-dependent l_α and r

We solve eq. 3 for r assuming that density-dependent, compensatory effects occur as changes in adult mortality change population size, but that this compensation affects preadult survival l_α only. We further assume that all females at and above a given age of maturity breed each year, although we recognize that variation in breeding at ages bracketing our set age at maturity may occur (see Walker 1994*a*, 1994*b*).

The compensation in preadult survival is assumed to occur as a response to population reduction, as in standard fisheries dynamics.

© 1997 NRC Canada

When fishing mortality (F) is applied to a population (assuming knife-edge selection at age α by the fishing gear with equal vulnerability at all subsequent ages), total adult mortality increases to Z (where $Z = M + F$). This decreases adult population size and therefore reproductive output. However, we are assuming here that the expected decrease in the population's intrinsic rate of increase r is at least partly compensated for by an increase in preadult survival l_α, which affects the level of r actually achieved. If this r level were known, the increased l_α level (designated $l_{\alpha,Z}$) could be found from Lotka's equation, which must remain satisfied in the long term. Notice that l_α is the net survival to age α, so we need not be concerned about the particular structure of the age-specific survival up to that age.

We can solve for this $l_{\alpha,Z}$ by setting $r = 0$. This enables us to find the preadult survival needed for a population to be in stationary equilibrium (neither increasing nor decreasing in time) under mortality Z and with fishing mortality F sustainable. Equation 3 with $r = 0$, mortality Z, and $l_\alpha = l_{\alpha,Z}$ then becomes

$$(4) \quad e^{-Z} + l_{\alpha,Z}b\,[1 - e^{-Z(w-\alpha+1)}] = 1.0,$$

from which $l_{\alpha,Z}$ can now be determined as a direct solution for the given level of Z.

The Z-reduced population with preadult survival $l_{\alpha,Z}$ is able to remain stationary and sustain the fishing mortality F because it then possesses a certain inherent and enhanced rate of increase r. This rate can be found by removing F, so that Z equals natural mortality M again, and determining how fast the population then grows. The growth would acquire a rate r (here designated r_Z) insofar as $l_{\alpha,Z}$ and the other parameters remain constant (possible if all surplus growth is shunted elsewhere, e.g., into a fishery). This r_Z would be a measure of the potential productivity at the Z-determined population size, and r_Z can be found from eq. 3 after substituting the $l_{\alpha,Z}$ estimate for l_α.

Rate of increase at the MSY population size

There are values of $l_{\alpha,Z}$ and r_Z for each Z level of mortality (assuming the required amount of l_α compensation is possible), but we are particularly interested in the $l_{\alpha,Z}$ and r_Z that characterize the population size producing maximum sustainable yield (MSY). We therefore set $Z = 2M$, the mortality level that should reduce a population to at least half its unexploited size. This is the size reduction commonly assumed to at least be required for producing MSY in many fish and other populations. Although such relationships have not been demonstrated for sharks, and their application could produce an overexploited condition (Thompson 1992), we will use $Z = 2M$ both to approximate the MSY condition and to standardize our computations (e.g., to allow comparison of r levels among shark species). Using eq. 4 with $Z = 2M$, $l_{\alpha,2M}$ is found and then substituted into eq. 3 to solve iteratively for $r = r_{2M}$, the productivity potential at the MSY population level.

Rate of increase and fecundity ratios

Although we assume that the fecundity or m_x schedule does not change, an allowance must be made for demographic effects on the arithmetic value of average fecundity b. For example, if r increases because of increased adult survival, mean fecundity b would increase as the demography shifts to more older fish, which have higher fecundity at age (m_x) values. However, if r increases enough, the faster growing population would become composed of proportionately more younger females, which would tend to lower mean fecundity b. Thus, b can be specified only if r is known, and while we can invoke $r = 0$ at the $Z = 2M$ population size, r_{2M} is an unknown as yet undetermined. However, it is the ratio of the b levels at these different r levels that affects r_{2M}. Thus, we can solve for r_{2M} by assuming reasonable b increases from the $r = 0$ condition, namely $1.00b$ (no increase), $1.25b$, or $1.50b$. These levels of b are substituted into eq. 3 along with $l_{\alpha,2M}$ to obtain a range of r_{2M} estimates. The higher factored b levels would apply to species that are more fecund.

Adjusting yield per recruit

The above estimates of intrinsic rate of increase (r_{2M}) are used to construct a Y/R determination that adjusts for the stock size dependent nature of shark recruitment. It is used to specify a stock–recruitment relationship that determines the amount of recruitment actually produced at each level of population or stock.

Stock–recruitment relationship

For a stock–recruitment (S–R) relationship, we use the normalized, one-parameter form of Beverton and Holt's (1957) asymptotic model. This S–R relationship can be written (see Ricker 1958, p. 269; 1975, p. 346) as

$$(5) \quad R' = P'/[1 - A(1 - P')]$$

where R' and P' are recruitment and stock fractional abundances, respectively, measured relative to their unexploited sizes (and therefore scaled from 0 to 1) and where A is a shape parameter (also varying from 0 to 1) governing the degree of dependence in the relationship. In terms of our discussion, a larger A means that for a given mortality Z, there is more compensation through increased preadult survival $l_{\alpha,Z}$.

Parameter A of eq. 5 can be determined by applying the r_{2M} estimate to the MSY exploitation relationship for that equation as given by Ricker (1975, p. 348):

$$(6) \quad \mu_s = 1 - (1 - A)^{1/2}$$

where μ_s is the rate of sustainable exploitation at the MSY population size. If this rate is estimated as the net rate of annual population increase, which here is $e^{r2M} - 1$, then substituting this rate for μ_s in eq. 6 allows direct solution for A. Once A is determined, the appropriate stock–recruitment relationship is specified and Y/R can be adjusted for recruitment changes resulting from different levels of fractional stock abundance.

Recruitment adjustment

Our interest being in adjusting a conventional Y/R analysis, we derived fractional stock abundance P' from the population biomasses at each level of fishing mortality (F) and age at first capture (t_r') as calculated in the ordinary Y/R analysis. As a measure of P', we expressed these biomasses as fractions of the biomass in the unfished state when $F = 0$. Then substituting these P' into eq. 5, recruitment level R' is found for each fishing condition. In this case, the abundance level P' is calculated from biomasses originally determined assuming constant recruitment.

The R' that is found is the level of recruitment relative to the level characterizing the unexploited state, and the recruitment-adjusted Y/R (designated Y/RA) is then simply

$$(7) \quad \text{Y/RA} = (\text{Y/R})R',$$

i.e., the per-recruit yield or productivity is adjusted for the new, fractional level of recruitment. Because R' is a dimensionless ratio, the dimensions of Y/RA are the same as for the original Y/R.

Parameters for the leopard shark

Life history parameters

The parameters needed to estimate the rate of increase for the leopard shark were derived, with some modification, from the published literature. Age at maturity (α) was reduced from 17 years (Cailliet

© 1997 NRC Canada

Table 2. Leopard shark life-history parameters and estimates of $l_{\alpha,2M}$ and r_{2M} at three levels of mean fecundity b.

α	w	M	$l_{\alpha,2M}$[a]	b	r_{2M}[b]
13	30	0.15	0.043	$1.00b = 6.02$	0.032
13	30	0.15	0.043	$1.25b = 7.53$	0.045
13	30	0.15	0.043	$1.50b = 9.03$	0.055

Note: Parameter units are as follows: α, years of age; w, years of age; M, per year; $l_{\alpha,2M}$, per year; b, female pups per adult female; r_{2M}, per year.
[a]From eq. 4.
[b]From using $l_{\alpha,2M}$ in eq. 3.

1992) to 13 years here, after considering the estimates of 10–15 years at maturity of Kusher et al. (1992) and Bane and Bane (1971) and also the sampling on which Cailliet's (1992) estimate of 17 years was based (Ackerman 1971). The maximum reproductive age (w) of females is 30 years as per Smith and Abramson (1990) and Cailliet (1992). The base level of average fecundity ($1.00b$) was set at 6.02 female pups per adult female (the abundance-weighted mean of m_x from age 13 onwards, calculated after extending the rising limb of Smith and Abramson's (1990) fecundity curve backwards from its value at age 19 to zero at age 12). Natural mortality ($M = 0.15$) was estimated from maximum reproductive age (w) as $\ln(M) = 1.44 - 0.982 \ln(30 \text{ years})$, as per Hoenig (1983).

Stock and recruitment levels

The fractional stock level P' for determining fractional recruitment level R' (eq. 5) was taken from Smith and Abramson's Y/R analysis of the leopard shark (Southwest Fisheries Science Center, P.O. Box 271, La Jolla, CA 92038, personal communication). Intermediate steps in their calculations enabled us to determine the biomasses per recruit at each age from age 4 to 30 years. We equated P' with their total exploited biomass expressed relative to their total unexploited biomass. Total exploited biomass was the summation of the biomasses at each age, without the effects of fishing mortality up to the age at first capture, and with fishing mortality thereafter. The levels of P' and R' were determined for each combination of fishing effort and age at first capture.

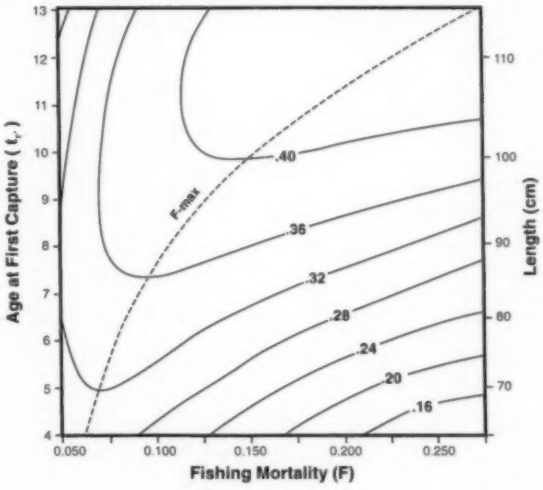
Results

Estimates of intrinsic rate of increase

The leopard shark's $l_{\alpha,Z}$ and r_Z were calculated for $Z = 2M$ (i.e., calculated for $l_{\alpha,2M}$ and r_{2M}) and for the three fecundities $1.00b$, $1.25b$, and $1.50b$ (Table 2). Because this shark is moderately fecund with litter size up to 18 female pups and fecundity increasing with female size (Ackerman 1971; Compagno 1984; Smith and Abramson 1990), these levels of b are possible. For this example, we optimistically took the average of the $1.25b$ and $1.50b$ values of r_{2M} as estimating the MSY level of r, i.e., we set $r_{2M} = 0.050$/year (see Table 2).

We also obtained r estimates (not to be confused with the r_{2M} estimates) using eq. 3, for comparison with estimates by the more typical life table method as found in the literature. Table 3 shows that when the same parameters are used in both methods, eq. 3 will give essentially the same value for r, save for rounding errors. Our r value for the leopard shark was 0.071/year. Notice that the estimates of r_{2M} in the previous Table 2, which are the r values at the MSY population size, involve additional, compensatory considerations, and so are not directly comparable with values of r in Table 3. Values

Fig. 1. Recruitment-adjusted yield per recruit (Y/RA) for the leopard shark.



from the literature may also reflect different sets of basic assumptions and input values. For example, the leopard shark r_{2M} in Table 2 is lower than the leopard shark r in Table 3, although the reverse would be expected because of the population compensation that is factored into the former. Most likely, this is because the r from the literature is calculated using different assumptions concerning juvenile survival.

Adjusted yield per recruit

With $r_{2M} = 0.050$/year, $e^{r_{2M}} - 1 = 0.051$/year was calculated as the leopard shark's net annual rate of increase at the MSY population level. Equating this rate to exploitation rate μ_x of eq. 6, parameter A of eq. 5 was determined to be 0.099. The fractional recruitment level R' from eq. 5 was thus $R' = P'/[1 - 0.099(1 - P')]$.

The R' values thus obtained were then used to adjust, by means of eq. 7, the Y/R values calculated by Smith and Abramson (1990) for the leopard shark . The resulting values of Y/RA are shown as a standard isopleth diagram (Fig. 1), along with Smith and Abramson's (1990) original, unadjusted Y/R (Fig. 2).

Discussion

Comparing the recruitment-adjusted Y/R pattern with the unadjusted pattern, it appears that the leopard shark may be even more susceptible to overexploitation than previously thought. Besides the much smaller magnitude of recruitment-adjusted Y/RA (Fig. 1) compared with unadjusted Y/R (Fig. 2), the main difference is that the isopleths of the former have a much greater horizontal trend over most of the Y/R field. At intermediate levels of fishing mortality, gains are therefore much greater from increasing the age at first capture than from increasing fishing effort. This means that the leopard shark is much easier to overfish than suggested by ordinary Y/R analysis, as shown by the strong shift to the left of the dashed loci of F-max in the Y/RA diagram (F-max is the F giving

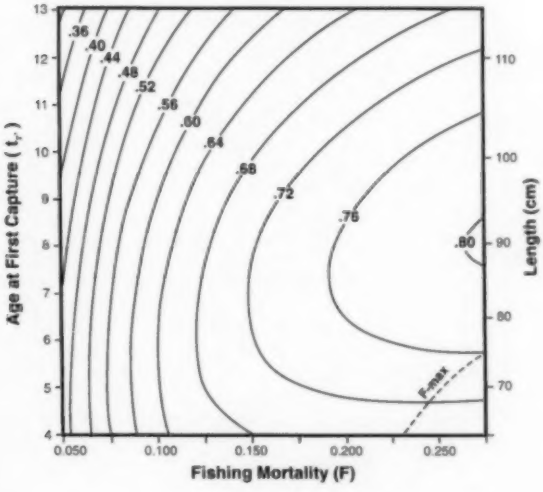
© 1997 NRC Canada

Table 3. Comparison of r estimates from the published literature and from applying eq. 3, using parameters as determined from the cited literature.

	Parameters					r estimates from:		
Shark species	α	w	b	M	l_α	literature	eq. 3	Source
Lemon (*Negaprion brevirostris*)	12	25	2.50	0.162	0.084	0.015	0.014	Hoenig and Gruber 1990
Sandbar (*Carcharhinus plumbeus*)	13	35	1.66	0.105	0.100	0.018	0.021	Hoff 1990
Atlantic sharpnose (*Rhizoprionodon terraenovae*)	4	10	2.49	0.420	0.186	0.044	0.045	Cortes 1995
Leopard (*Triakis semifasciata*)	17	30	8.44	0.140	0.080	0.067	0.071	Bane and Bane 1971; Cailliet 1992; Kusher et al. 1992
Dogfish (*Squalus acanthias*)	29	69	1.55	0.094	0.760	0.077	0.071	Jones and Geen 1977
Angel (*Squatina californica*)	10	35	3.00	0.100–0.200	0.368–0.135	0.056–0.149[a]	0.058–0.158	Cailliet et al. 1992; Natanson and Cailliet 1986

[a]Upper r estimate recalculated from original $r = 0.136$/year.

Fig. 2. Standard yield per recruit (Y/R) for the leopard shark as determined by Smith and Abramson (1990).



maximum Y/R at any given age at first capture). These results stem from the low intrinsic rate of population increase and therefore poor compensatory capability of this as well as many other sharks.

The leopard shark population we describe was fished primarily by angling (Smith and Abramson 1990), and so our assumption of an equal rate of fishing mortality on all adults and, for the Y/R analysis, on all those above the age at first capture is tenable. For species or stocks exploited by highly size-selective gear, such as gill nets (Kirkwood and Walker 1986), similar analyses would require specifying a varying age-specific mortality schedule. Nevertheless, our results are in line with experience and intuitive understanding of how slow-growing, low-fecundity fishes should be exploited, i.e., the rate of exploitation should be low and the age at first capture high.

We have shown how even limited information consisting basically of only the age at first maturity, maximum reproductive age, and average fecundity can, when combined with simple concepts of density-dependent compensation from standard population dynamics, be expanded to obtain important life history information for use in guiding fisheries management (of course, the parameters we used for the leopard shark to illustrate and test our method came from the detailed life history data available for that species). The information was used to show how lowered recruitment from stock depletion reduces productivity of the leopard shark stock and the level of exploitation that would be prudent. Such considerations as these that emphasize cautious exploitation are in the spirit and direction that would derive from a "return to population biology," as recommended by R.J.H. Beverton (Langstone, Gwent NO6 2JU, U.K., personal communication, September 1994).

Other researchers have also incorporated density-dependent compensation using age-structured, dynamic simulation modelling of shark populations. In such a study, Wood et al. (1979) concluded that density dependence is most likely mediated through changes in natural mortality, but they set this mortality as being at the same rate for all ages. Our study differs in that density-dependent compensation is effected mainly through changes in juvenile survival. Walker (1992, 1994*a*, 1994*b*) likewise incorporated density-dependent natural mortality into his fully age-structured models and showed how increased survival among prerecruits can compensate for the effects of reduced parental stock size upon recruitment. He separately examined compensatory effects on prerecruits only (Walker 1992, 1994*a*), all ages (Walker 1994*a*), and all ages but with greatest effect on prerecruits and senescent fish (Walker 1994*b*). Our approach is not meant to replace these more expanded and comprehensive models, but to serve as a secondary option when the full range of data are lacking.

In conclusion, our main purpose is to show how population productivity (r) of sharks can be estimated using a demographic procedure that overcomes the usual lack of information on preadult survival rates. This is accomplished by invoking standard concepts from fisheries population

© 1997 NRC Canada

dynamics, and by calculating a range of possible r (actually, r_{2M}) to allow for imprecision. Our procedure can also reduce arbitrariness in estimating survival to specific ages (l_x), which, in turn, can more tightly contain the range of r estimates. Furthermore, by determining values of r at certain levels of adult mortality standardized according to some rationale, e.g., at $Z = 2M$, between-species comparisons are made more meaningful. This method should prove useful for estimating and comparing productivities among shark species for which there are few empirical observations and for species not normally considered amenable to standard demographic analysis.

Acknowledgements

We profited greatly from discussing the ideas presented here with Patrick K. Tomlinson (Inter-American Tropical Tuna Commission, La Jolla, Calif.), Terry I. Walker (Victorian Fisheries Research Institute, Queenscliff, Australia), the late Professor Raymond J.H. Beverton (Langstone, United Kingdom), and Gregor Cailliet (Moss Landing Marine Laboratories, California); we also benefited from valuable comments given by the *Journal* referees.

References

Ackerman, L.T. 1971. Contribution to the biology of the leopard shark, *Triakis semifasciata* (Girard) in Elkhorn Slough, Monterey Bay, California. M.A. thesis, Sacramento State College, Sacramento, Calif.

Bane, G.W., and Bane, A.W. 1971. Bay fishes of northern California. Mariscos Publ., Southhampton, N.Y.

Beverton, R.J.H., and Holt, S.J. 1957. On the dynamics of exploited fish populations. Fish. Invest. Ser. II Mar. Fish. G.B. Minist. Agric. Fish. Food Vol. 19.

Cailliet, G.M. 1992. Demography of the central California population of the leopard shark (*Triakis semifasciata*). Aust. J. Mar. Freshwater Res. **43**: 183–193.

Cailliet, G.M., Mollet, H.F., Pittenger, G.G., Bedford, D., and Natanson, L.J. 1992. Growth and demography of the Pacific angel shark (*Squatina californica*), based upon tag returns off California. Aust. J. Mar. Freshwater Res. **43**: 1313–1330.

Compagno, L.J.V. 1984. Sharks of the world: an annotated and illustrated catalogue of shark species known to date. *In* FAO species catalogue. Vol. 4. Part 2. Carcharhiniformes. FAO Fish. Synop. **125**: 251–655.

Cortes, E. 1995. Demographic analysis of the Atlantic sharpnose shark *Rhizoprionodon terraenovae*, in the Gulf of Mexico. Fish. Bull. **93**: 57–66.

Hoenig, J.M. 1983. Empirical use of longevity data to estimate mortality rates. Fish. Bull. **81**: 898–903.

Hoenig, J.M., and Gruber, S.H. 1990. Life history patterns in the elasmobranchs: implications for fisheries management. *In* Elasmobranchs as living resources: advances in the biology, ecology, systematics, and the status of the fisheries. *Edited by* H.L. Pratt, Jr., S.H. Gruber, and T. Taniuchi. NOAA Tech. Rep. NMFS No. 90. pp. 1–16.

Hoff, T.B. 1990. Conservation and management of the western North Atlantic shark resource based on the life history strategy limitations of the sandbar shark. Ph.D. dissertation, University of Delaware, Newark, Del.

Holden, M.J. 1973. Are long-term sustainable fisheries for elasmobranchs possible? Rapp. P.-V. Réun. Cons. Int. Explor. Mer, **164**: 360–367.

Jones, B.C., and Green, G.H. 1977. Reproduction and embryonic development of spiny dogfish (*Squalus acanthias*) in the Strait of Georgia, British Columbia. J. Fish. Res. Board Can. **34**: 1286–1292.

Kimura, D.K. 1988. Stock–recruitment curves as used in the stock-reduction analysis model. J. Cons. Int. Explor. Mer, **44**: 253–258.

Kirkwood, G.P., and Walker, T.I. 1986. Gill net mesh selectivities for gummy shark, *Mustelus antarticus* Günther, taken in southeastern Australian waters. Aust. J. Mar. Freshwater Res. **37**: 689–697.

Kusher, D.I., Smith, S.E., and Cailliet, G.M. 1992. Validated age and growth of the leopard shark, *Triakis semifasciata*, with comments on reproduction. Environ. Biol. Fishes, **35**: 187–203.

Lotka, A.J. 1959. Principles of mathematical biology. (Reprinted edition.) Dover, New York.

Natanson, L.J., and Cailliet, G.M. 1986. Reproduction and development of the Pacific angel shark, *Squatina californica*, off Santa Barbara, California. Copeia, **4**: 987–994.

Ricker, W.E. 1958. Handbook of computations for biological statistics of fish populations. Bull. Fish. Res. Board Can. No. 119.

Ricker, W.E. 1975. Computation and interpretation of biological statistics of fish populations. Bull. Fish. Res. Board Can. No. 191.

Smith, S.E., and Abramson, N.J. 1990. Leopard shark *Triakis semifasciata* distribution, mortality rate, yield, and stock replenishment estimates based on a tagging study in San Francisco Bay. Fish. Bull. **88**: 371–381.

Stearns, S.C. 1992. The evolution of life histories. Oxford University Press, New York.

Thompson, G.G. 1992. Management advice from a simple dynamic pool model. Fish. Bull. **90**: 552–560.

Walker, T.I. 1992. Fishery simulation model for sharks applied to the gummy shark, *Mustelus antarticus* Günther, from southern Australian waters. Aust. J. Mar. Freshwater Res. **43**: 195–212.

Walker, T.I. 1994*a*. Stock assessments of the gummy shark, *Mustelus antarcticus* Günther, in Bass Strait and off South Australia. *In* Population dynamics for fisheries management. *Edited by* D.A. Hancock. Australian Society for Fish Biology Workshop Proceedings, Perth, 24–25 August 1993. Australian Society for Fish Biology, Perth, Australia. pp. 173–187.

Walker, T.I. 1994*b*. Fishery model of gummy shark, *Mustelus antarcticus*, for Bass Strait. *In* New opportunities: best practice. *Edited by* I. Bishop. Resource Technology '94 Conference Proceedings, Melbourne, 26–30 September 1994. The Center for Geographic Information Systems and Modelling, University of Melbourne, Melbourne, Australia. pp. 422–438.

Wood, C.C., Ketchen, K.S., and Beamish, R.J. 1979. Population dynamics of spiny dogfish (*Squalus acanthias*) in British Columbia waters. J. Fish. Res. Board Can. **36**: 647–656.

© 1997 NRC Canada

Evidence for natural hybridization between Dolly Varden (*Salvelinus malma*) and bull trout (*Salvelinus confluentus*) in a northcentral British Columbia watershed

James S. Baxter, Eric B. Taylor, Robert H. Devlin, John Hagen, and J. Donald McPhail

Abstract: Morphological and molecular analyses were used to identify sympatric Dolly Varden (*Salvelinus malma*) and bull trout (*Salvelinus confluentus*) and their hybrids from streams tributary to Thutade Lake, Peace River drainage, northcentral British Columbia. Morphological analysis identified two groups of char resembling allopatric Dolly Varden and bull trout as well as a small number of morphologically intermediate fish. One of the morphologically intermediate char possessed both Dolly Varden and bull trout ribosomal DNA (rDNA) and growth hormone restriction fragment length differences (RFLDs) indicating its F_1 hybrid status. Another morphologically intermediate fish exhibited a hybrid rDNA RFLD, but possessed a growth hormone RFLD consistent with a hybrid × hybrid mating or a hybrid × bull trout backcross. Both hybrid individuals had a diagnostic bull trout mitochondrial DNA (mtDNA) RFLD. As mtDNA is maternally inherited, our data imply that the F_1 hybrid resulted from a mating between a bull trout female and a Dolly Varden male, perhaps by small Dolly Varden males "sneaking" fertilizations during bull trout spawnings. This is the first record of Dolly Varden in the Peace River drainage, and the first confirmation of natural hybrids between the two species. Despite apparent introgression, the two species appear to be maintaining themselves as distinct genomes.

Résumé : Des analyses morphologiques et moléculaires ont été employées pour identifier des Dolly Varden (*Salvelinus malma*) et des ombles à tête plate (*Salvelinus confluentus*) sympatriques et leurs hybrides dans des cours d'eau tributaires du lac Thutade, bassin de la rivière de la Paix, dans le centre-nord de la Colombie-Britannique. L'analyse morphologique a identifié deux groupes d'ombles ressemblant aux Dolly Varden et ombles à tête plate allopatriques de même qu'un petit nombre de poissons morphologiquement intermédiaires. Un des ombles morphologiquement intermédiaires possédait l'ADN ribosomique (ADNr) et des différences de longueurs des fragments de restriction (DLFR) pour l'hormone de croissance appartenant à la Dolly Varden et à l'omble à tête plate, ce qui indiquait son état d'hybride F_1. Un autrepoisson morphologiquement intermédiaire présentait des DLFR de l'ADNr hybride, mais possédait une DLFR pour l'hormone de croissance concordant avec un croisement hybride–hybride ou un rétrocroisement hybride–omble à tête plate. Les deux individus hybrides possédaient une DLFR de l'ADN mitochondrial (ADNmt) indicatifs de l'omble à tête plate. Comme l'ADNmt est hérité de la femelle, nos données laissent entendre que l'hybride F_1 provient d'un croisement entre une femelle d'omble à tête plate et un mâle Dolly Varden, peut-être par suite d'une fécondation « insidieuse » par des mâles Dolly Varden de petite taille durant le frai des ombles à tête plate. Il s'agit du premier signalement de Dolly Varden dans le bassin hydrographique de la rivière de la Paix et la première confirmation de l'existence d'hybrides naturels entre ces deux espèces. En dépit d'une introgression apparente, les deux espèces semblent vouloir se maintenir comme des génomes distincts.
[Traduit par la Rédaction]

Introduction

Dolly Varden (*Salvelinus malma*) and bull trout (*Salvelinus confluentus*) are native char that are widely distributed in western North America. Until recently, their taxonomic status was confused, and bull trout were considered conspecific with Dolly Varden (e.g., McPhail 1961; Morton 1970). In 1978, however, Cavender demonstrated that the bull trout is a species separate from Dolly Varden, and recent cytological and molecular research (Cavender 1984; Phillips et al. 1989; Grewe et al. 1990; Pleyte et al. 1992) has strongly supported this contention. Nonetheless, most of the morphological and molecular data used to support the bull trout's specific status come from regions where Dolly Varden do not occur. Yet, in char, as in many other fish, the ultimate test of specific status is the ability of putative species to coexist. Consequently, what happens in sympatry is critical to our understanding of the ecological and genetic relationships of Dolly Varden and bull trout, and in these species, the area of sympatry appears to be confined to British Columbia and western Washington

Received March 12, 1996. Accepted July 16, 1996.
J13349

J.S. Baxter, E.B. Taylor,[1] J. Hagen, and J.D. McPhail. Department of Zoology, University of British Columbia, 6270 University Boulevard, Vancouver, BC V6T 1Z4, Canada.
R.H. Devlin. Department of Fisheries and Oceans, West Vancouver Laboratory, West Vancouver, BC V7V 1N6, Canada.

[1] Author to whom correspondence should be addressed.
e-mail: etaylor@zoology.ubc.ca

© 1997 NRC Canada

Fig. 1. Distribution of Dolly Varden and bull trout in British Columbia showing areas of actual or potential overlap relative to the Thutade Lake watershed.

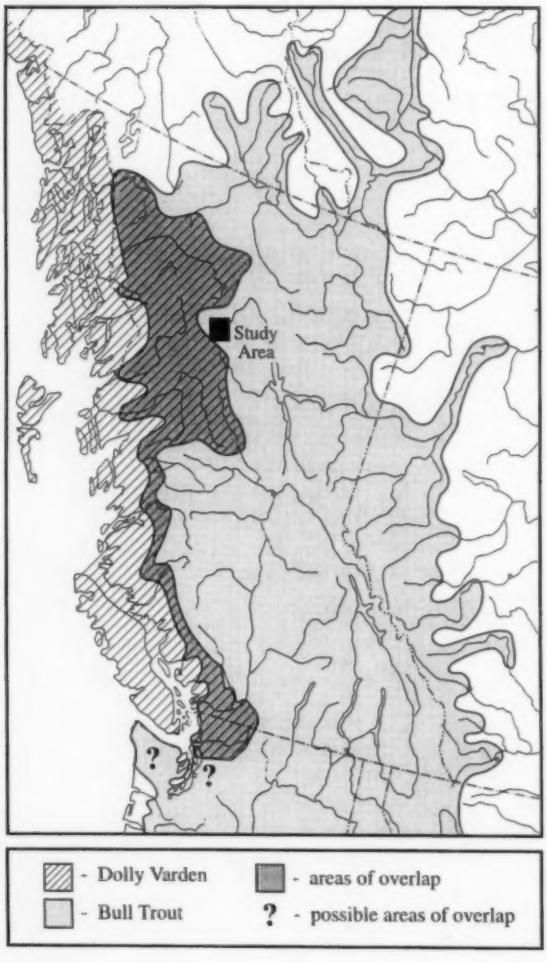
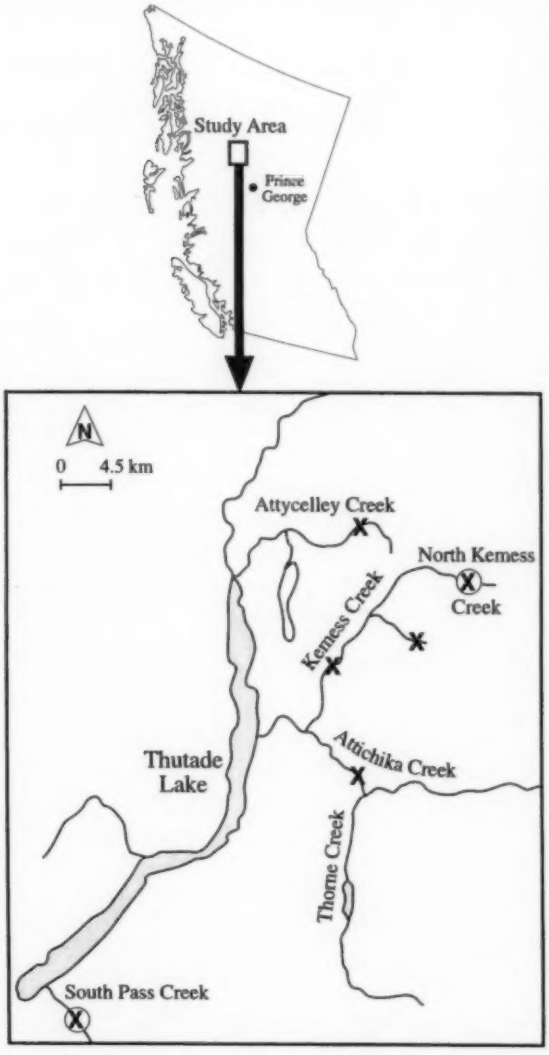


Fig. 2. Thutade Lake study area showing sample sites (crosses) and locations where hybrid char were found (encircled crosses).



(Fig. 1). This report is the first in a series of studies on the genetic and ecological relationships between sympatric Dolly Varden and bull trout.

Haas and McPhail (1991) examined Dolly Varden and bull trout throughout their North American ranges and developed a linear discriminant function (LDF) based on four morphometric and meristic traits. The LDF can be used to classify individual fish as either Dolly Varden or bull trout. In areas where the two species coexist, however, identification is not straightforward and individual char often have intermediate LDF scores (e.g., Brown 1992). The presence of morphologically intermediate individuals in sympatry raises the possibility of hybridization between Dolly Varden and bull trout. Indeed, other authors (Cavender 1978; Haas and McPhail 1991; Markle 1992; Phillips et al. 1994) have suggested the possibility of hybridization between the species, but there are no confirmed records of such hybrids and no estimates of either the level of hybridization or whether there is gene flow between the species. To clarify the morphology of hybrids, Haas and McPhail (1991) produced artificial F_1 hybrids between Dolly Varden and bull trout and demonstrated that they were morphologically intermediate to the parental species.

In this study, we document the presence of sympatric populations of Dolly Varden and bull trout in the Thutade Lake drainage in northcentral British Columbia and demonstrate the occurrence of natural hybrids and backcrosses in this pristine environment.

Materials and methods

Study sites

The study area consists of streams tributary to Thutade Lake in northcentral British Columbia (Fig. 2). Thutade Lake is in the

© 1997 NRC Canada

southwestern headwaters of the Mackenzie River system and connects to the Mackenzie via the upper Finlay and Peace river systems. The study sites were located on Attichika (and its tributary, Kemess Creek), Attycelley, and South Pass creeks (all tributary to Thutade Lake).

Char collections

Char samples were collected from each study site by electroshocking in August–September 1994. For morphological analysis, char (N = 74) were preserved in 10% formalin for 2 weeks, soaked in water for 2 days, and then stored in 40% isopropanol. Blood, liver, and fin tissues were collected from a subsample of the char examined for morphological analysis (N = 33) and were stored in 95% ethanol at 4°C.

Morphological analysis

Among the morphological and meristic characters studied by Haas and McPhail (1991), total branchiostegal ray number, anal fin ray number, and the ratio of upper jaw length to standard length can identify individuals of the two species from allopatric populations with 100% accuracy using LDF analysis. Differences between the species in these traits are inherited, and artifical hybrids between them are morphologically intermediate; Dolly Varden have fewer branchiostegal and anal fin rays and a lower ratio of upper jaw length to standard length than do bull trout (Haas and McPhail 1991). We scored these traits in our char samples from the Thutade Lake watershed to obtain evidence of sympatric populations of char and of hybrids between them on the basis of the characterization of the species and their hybrids made by Haas and McPhail (1991). The measurements were made using vernier calipers, and the counts were done under a dissecting microscope.

As recommended by Neff and Smith (1979), we used principal components (PC) analysis of the data correlation matrix (Pimentel 1979) to produce a multivariate summary score of morphological variation between the species and to obtain evidence for hybrids. Given the known morphological differences between allopatric Dolly Varden and bull trout, a bimodal distribution of PC scores would be evidence of sympatric populations with hybrid PC scores occupying an intermediate position (cf. Neff and Smith 1979; Campton 1987).

Molecular analyses

We used three assays of molecular genetic variation in char DNA to examine the question of hybridization between sympatric Dolly Varden and bull trout. Grewe et al. (1990) resolved eight mitochondrial DNA (mtDNA) restriction site differences with six enzymes to produce species-specific restriction site profiles for Dolly Varden and bull trout. We used the enzyme *Hin*dIII to distinguish Dolly Varden from bull trout mtDNA. The *Hin*dIII dimorphism results from a single restriction site change between the two species (five sites in Dolly Varden and four sites in bull trout). Extraction of DNA from ethanol-stored tissue followed the pronase, or proteinase K, digestion and phenol–chloroform extraction procedures outlined in Taylor et al. (1996). Genomic DNA samples (5 μg) were digested with *Hin*dIII (New England Biolabs), electrophoresed in 1.0% agarose gels, and Southern blotted to nylon membranes as described in Taylor et al. (1996). Restriction site variation was then assayed using hybridization of membrane-bound char DNA with digoxigenin-labelled rainbow smelt (*Osmerus mordax*) mtDNA at 58°C followed by detection of probe-char DNA hybrids by chemiluminescence (Taylor et al. 1996). In our inital sampling, we also assayed restriction site variation between the species with two other diagnostic enzymes, *Pvu*II and *Bam*HI. Restriction site differences between the species evident at *Hin*dIII were always associated with differences at these other sites. Consequently, to identify either Dolly Varden or bull trout mtDNA, we assayed the bulk of our samples for the *Hin*dIII site difference. As mtDNA is maternally inherited (Avise and Lansman 1983), this analysis detects the presence of one, or the other, species' mtDNA in each sample, but cannot detect hybrids. If combined with an analysis of a biparentally inherited marker (see below), however, mtDNA characterization of hybrids can reveal the directionality of hybridization (i.e., the specific identity of the female in the hybrid mating).

The second molecular marker involved restriction site analysis of the first internal transcribed spacer region of the ribosomal DNA (rDNA) gene complex. Ribosomal DNA is a class of tandemly repetitive nuclear DNA consisting of coding and noncoding (spacer regions) DNA sequences. The coding regions produce rRNAs that combine with ribosomal proteins to produce ribosomes (reviewed in Hillis and Dixon 1991). In a phylogenetic reconstruction within the genus, Pleyte et al. (1992) sequenced 600 base pairs (bp) of the first internal transcribed spacer region (ITS 1) isolated from six species of *Salvelinus*. Dolly Varden and bull trout differed in their ITS 1 sequences by approximately 8.6% (Pleyte et al. 1992) and, interestingly, were not each other's closest relative; Dolly Varden were most closely aligned with Arctic char (*Salvelinus alpinus*, 1.3% difference) whereas bull trout were most closely related to Japanese char (*Salvelinus leucomaenis*, 6.0% difference).

Comparison of the published ITS 1 sequences of Dolly Varden and bull trout indicated several restriction enzymes that would reveal restriction site differences between the species. To exploit these differences, we used the polymerase chain reaction (PCR) to amplify the ITS 1 region from fish using primers found in the flanking 18S and 5.8S coding regions. The primers used in the PCR reactions were 5′ AAA AAG CTT TTG TAC ACA CCG CCC GTC GC 3′ (ITS 1 forward) and 5′ AGC TTG CTG CGT TCT TCA TCG A 3′ (ITS 1 reverse) as described in Pleyte et al. (1992). PCR reactions were run in 40-μL reaction volumes under the following conditions (final concentrations): 1 μg of template, 1.5 U of *Taq* polymerase (Promega), 3 mM $MgCl_2$, dATP, dTTP, dCTP, and dGTP each at 200 μmol, 350 pmol of each primer, 1% Triton X-100, 10 mM Tris–HCl (pH 9.0), and 50 mM KCl. In addition, 1 U of Perfect Match enzyme (Stratagene) was added to each reaction to reduce the amplification of nontarget DNA fragments (cf. Pleyte et al. 1992). Amplification reactions were carried out under the following cycling conditions: 30 cycles of denaturation at 94°C for 1 min, annealing at 55°C for 1 min, and primer extension at 72°C for 1.5 min, followed by a single cycle of extension at 72°C for 5 min.

As predicted from their published sequences (Pleyte et al. 1992), when the ITS 1 PCR products were incubated with a number of enzymes, restriction site differences between Dolly Varden and bull trout were revealed. One of these enzymes, *Sma*I, cuts the 825-bp amplification product of bull trout into two smaller fragments (500 and 325 bp), but the 825-bp fragment is not cut in Dolly Varden. As rDNA is biparentally inherited and is confined to a single chromosomal location in Dolly Varden and bull trout (Phillips et al. 1989), first-generation hybrids between the species will produce a composite *Sma*I restriction fragment length difference. That is, pure Dolly Varden will have a single 825-bp fragment, pure bull trout will have two fragments (500 and 325 bp), and hybrids will have three fragments (825, 500, and 325 bp). One potential problem with this technique is that incomplete digestion of PCR products with *Sma*I could be mistaken for a hybrid pattern. We eliminated this potential problem by (*i*) cutting PCR products with several other enzymes that distinguish the two sequences (e.g., *Ava*I, *Sty*I, *Rsa*I), (*ii*) incubating reactions in the presence of excess enyzme in larger reaction volumes, and (*iii*) performing all enzyme incubations over at least 16 h (i.e., overnight). We never experienced any partial digestion artifacts, and in all cases, hybrid fish identifications were corroborated using enzymes other than *Sma*I.

PCR products (2 μL) were incubated with 20 units of the appropriate enzyme (New England Biolabs or Boehringer Mannheim). Restricted PCR products were electrophoresed in 2% agarose gels and the molecular masses of fragments were estimated by comparison with molecular mass size standards (BRL

© 1997 NRC Canada

Table 1. Location and sample sizes for allopatric Dolly Varden and bull trout examined for mtDNA, rDNA, and GH RFLD variation.

Species	Population	Watershed	*N*		
			mtDNA	rDNA	GH
S. malma	Cowichan Lake	Vancouver Island	5	5	5
	Cub Creek	Vancouver Island	1	1	1
	Keogh River	Vancouver Island	1	1	1
	Misty Lake	Vancouver Island	1	1	1
	Upper Brent Creek	Queen Charlotte Islands	5	5	5
	Honna River	Queen Charlotte Islands	3	3	5
	Aero River	Queen Charlotte Islands	3	3	3
	Kumealon Lake	Central B.C. coast	3	3	3
	Unnamed Creek	Central B.C. coast	1	1	1
	Ayton Creek	Lower Skeena River	2	2	2
Total			25	25	27
S. confluentus	Metolius River	Lower Columbia River	11	3	4
	Squamish River	South coast B.C.	2	2	2
	Pitt Lake	Lower Fraser River	6	3	1
	Chilliwack Lake	Lower Fraser River	2	2	2
	Dominion Creek	Upper Fraser River	10	3	5
	Nahatlach River	Upper Fraser River	5	2	5
	Marshall Creek	Upper Fraser River	1	1	1
	Upper Arrow Lake	Upper Columbia River	14	6	10
	Mud Lake	Upper Fraser River	4	4	4
	North Thompson River	Upper Fraser River	4	4	4
	Tum Tum Lake	Upper Fraser River	3	3	3
Total			62	33	41

1-kb ladder) that included fragments ranging in size from 200 to 1000 bp.

Our third molecular marker was allelic variation at the two growth hormone (GH) loci using a cDNA probe developed from a sockeye salmon (*Oncorhynchus nerka*) GH gene (Devlin 1993). The cDNA resolves restriction fragment length differences (RFLDs) at two loci simultaneously (GH1 and GH2) in a variety of salmonid species (R.H. Devlin, unpublished data). Variation at this duplicated locus was assayed by Southern hybridization of *Hin*cII-restricted genomic DNA. Between 25 and 30 ng of the GH cDNA was labelled with [α-^{32}P]dCTP by random priming (Feinberg and Vogelstein 1983). Hybridizations were carried out in 5 × SSC and 5× Denhardt's solution (0.02% bovine serum albumin, 0.02% Ficoll, 0.02% polyvinylpyrrolidone) at 60°C in the presence of 0.1% calf thymus as competitor. Final stringency washes were 1% SSC –0.1% SDS at 60°C. When assayed in this way, Dolly Varden and bull trout are homozygous for alternative alleles at the two GH loci. At GH1, all Dolly Varden assayed to date (see below) exhibit a single allele of 2.9 kilobase pairs (kb) whereas bull trout exhibit a 3.5-kb allele. At GH2, Dolly Varden exhibit a 2.5-kb allele whereas bull trout possess a 2.2-kb allele.

The complementary features of mtDNA and nuclear-encoded markers have been exploited to study the extent, and directionality, of interspecific hybridization in several other species (e.g., Baker et al. 1989; Herke et al. 1990; Bernatchez et al. 1995). All such analyses assume the absence of within-species variation for the markers employed. In our case, the published mtDNA (Grewe et al. 1990) and rDNA (Pleyte et al. 1992) species distinctions were based on limited geographic sampling of populations within species. To confirm the absence of within-species variation in our molecular markers, we examined samples of "pure" allopatric Dolly Varden and bull trout from a series of widely scattered localities (Table 1): 10 Dolly Varden populations ranging from southern Vancouver Island to the Queen Charlotte Islands and 11 bull trout populations distributed from the upper Fraser River to the Metolius River, in Oregon. In over 100 allopatric char assayed, there was no evidence of within-species variation in the mtDNA, rDNA, and GH markers. Thus, we conclude that they are diagnostic for each of the species. We also confirmed the maternal inheritance of the mtDNA RFLDs, and the biparental inheritance of rDNA ITS 1 and GH RFLDs, respectively, using DNA from some of the laboratory-raised hybrids produced by Haas and McPhail (1991).

Results

Morphological analysis

Morphological analysis of char collected in the Thutade Lake watershed indicated that both Dolly Varden and bull trout were present throughout the study area. PC analysis of the three characters summarized 71% of the variation along the first axis (Table 2), which separated the specimens into two main groups (Fig. 3). The first group had a modal PC 1 score of −0.625 and corresponded to fish with a low number of branchiostegal and anal fin rays and a low ratio of upper jaw length to standard length. The second group of fish had a modal PC 1 score of 1.2 and these fish were characterized by higher values for all three traits. The presence of two main morphological groups of char, and the distinctions between them, are consistent with morphological characterizations of allopatric Dolly Varden and bull trout (Fig. 3; cf. Haas and McPhail 1991). The two modal groups of char were linked by

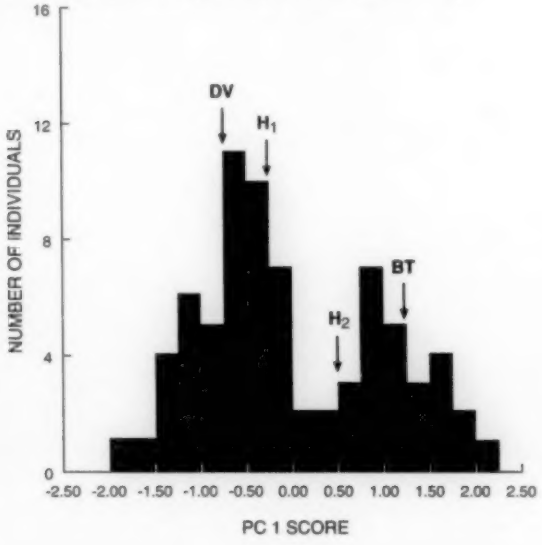
© 1997 NRC Canada

Table 2. Component score coefficients for a PC analysis of char from the Thutade Lake watershed.

Trait	PC 1	PC 2	PC 3
BRN	0.429	0.023	−1.441
AFRN	0.383	0.899	0.846
UPJ:SL	0.379	−0.934	0.778
% of total variance	70.6	19.5	9.8

Note: BRN, branchiostegal ray number; AFRN, anal fin ray number; and UPJ:SL, ratio of upper jaw length to standard length. Also given is the percentage of total morphological variation that is accounted for by each component axis.

Fig. 3. Histogram of standardized scores along PC axis 1 for char collected from the Thutade Lake watershed. The analysis was based on 74 char scored for number of branchiostegal rays (BRN), number of anal fin rays (AFRN), and the ratio of upper jaw length to standard length (UPJ:SL). Arrows indicate the PC 1 score for char with values of BRN, AFRN, and UPJ:SL equal to the medians for allopatric Dolly Varden (DV) and bull trout (BT), respectively, reported by Hass and McPhail (1991) or the PC 1 scores for the F_1 (H_1) and post-F_1 (H_2) hybrids, respectively, from the present study.

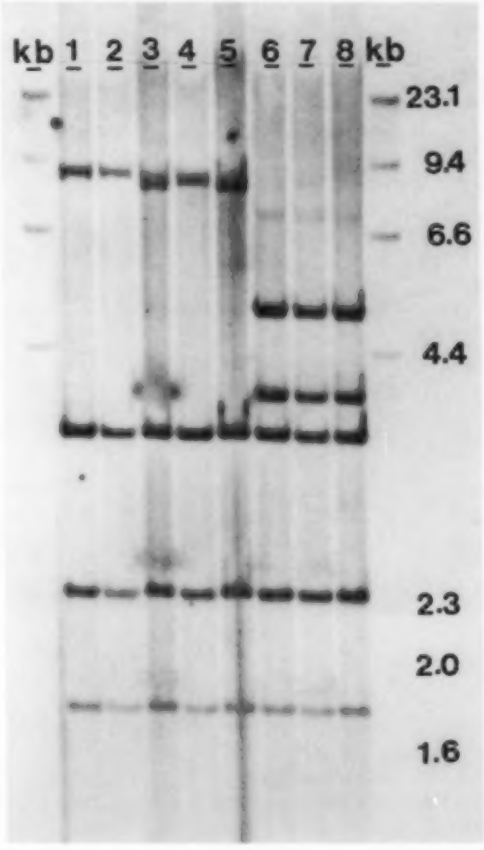


a smaller number of fish ($N = 9$) with intermediate PC 1 scores (arbitrarily defined as −0.25 to 0.50, Fig. 3). Histograms of scores along PC 2 and PC 3 (19% and 10 of total variation, respectively) were unimodal (data not shown).

Molecular variation

A total of 33 char for which morphological data were taken were also available for molecular analysis. Of the 33 fish examined for the *Hin*dIII mtDNA restriction site difference, 20 carried Dolly Varden mtDNA and 13 carried bull trout mtDNA (e.g., Fig. 4). All fish with Dolly Varden mtDNA had PC 1 scores below 0 (mean ± SE = −0.673 ± 0.07, $N = 20$). All but one fish with bull trout mtDNA had PC 1 scores greater than 0 (1.18 ± 0.09, $N = 13$).

Fig. 4. Mitochondrial DNA restriction fragment haplotypes used to distinguish bull trout and Dolly Varden. Haplotypes were resolved by restricting genomic DNA with *Hin*dIII followed by hybridization with digoxigenin-labelled *O. mordax* mtDNA at 58°C. Bull trout: lane 1, Metolius River (Oregon); lane 2, North Thompson River (upper Fraser River, B.C.); lane 3, Attycelley Creek (Thutade Lake, B.C.); lane 4, South Pass Creek (Thutade Lake, B.C.); lane 5, North Kemess Creek (Thutade Lake, B.C.). Dolly Varden: lane 6, Attycelley Creek (Thutade Lake, B.C.); lane 7, Cowichan Lake (Vancouver Island, B.C.); lane 8, Honna River (Queen Charlotte Islands, B.C.). kb, molecular mass size marker.



The 33 char assayed for mtDNA variation were also assayed for *Sma*I restriction site variation of the PCR-amplified ITS 1 fragment and the *Hin*cII site variation at the GH loci. Twenty-two char had the ITS 1 and GH RFLDs diagnostic of Dolly Varden and nine had RFLDs diagnostic of bull trout (Figs. 5 and 6). All of the char with nuclear markers diagnostic of Dolly Varden also possessed Dolly Varden mtDNA whereas all fish with bull trout nuclear markers carried bull trout mtDNA. Two fish, however, had three-banded rDNA patterns corresponding to fragments diagnostic of both species (lanes 5 and 6, Fig. 5). One of these fish was heterozygous for Dolly Varden and bull trout GH RFLDs at both loci, comfirming its status as an F_1 hybrid (lane 3, Fig. 6). The other char with a three-banded rDNA phenotype (lane 6,

© 1997 NRC Canada

Fig. 5. Ribosomal DNA genotypes used to distinguish Dolly Varden, bull trout, and their hybrids. Genotypes were resolved by cutting PCR-amplified char ITS 1 rDNA with *Sma*I and electrophoresing samples in 2.0% agarose gels. Dolly Varden: lane 1, Cowichan Lake (Vancouver Island, B.C.); lane 2, Honna River (Queen Charlotte Islands, B.C.); lane 3, Kumealon Lake (central B.C. coast); lane 4, Attycelley Creek (Thutade Lake, B.C.). Hybrid char: lane 5, South Pass Creek (Thutade Lake, B.C.); lane 6, North Kemess Creek (Thutade Lake, B.C.); Bull trout: lane 7, Attycelley Creek (Thutade Lake, B.C.); lane 8, Metolius River (Oregon); lane 9, North Thompson River (upper Fraser River, B.C.). Thutade Lake individuals in lanes 4–7 are the same as those in lanes 6, 5, 4, and 3, respectively, in Fig. 4. kb, molecular mass size marker.

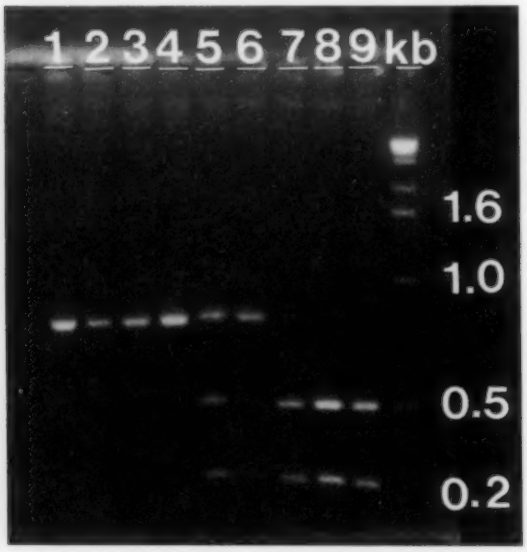


Fig. 6. Growth hormone genotypes used to distinguish bull trout, Dolly Varden, and their hybrids. Genotypes were resolved by hybridization of *Hinc*II-restricted genomic DNA with a sockeye salmon (*O. nerka*) GH cDNA at 60°C. Lane 1, bull trout; lane 2, Dolly Varden; lane 3, F_1 hybrid; lane 4, post-F_1 hybrid or backcross. Approximate molecular masses (kb) are given on the left.

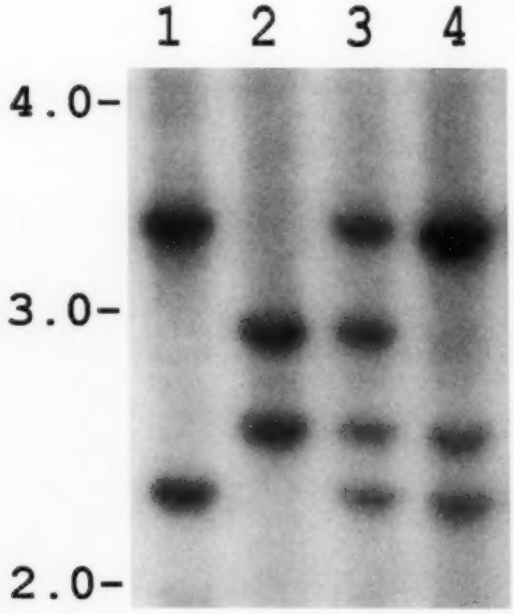


Fig. 5) exhibited differential intensity of the ethidium bromide stained RFLDs; the higher molecular mass fragment diagnostic of Dolly Varden was more intense than the two lower molecular mass bull trout fragments. This char also possessed a recombinant GH RFLD; it was homozygous for the bull trout GH1 allele, but heterozygous for Dolly Varden and bull trout restriction fragments at GH2, indicating that it is a post-F_1-generation hybrid or backcross (lane 4, Fig. 6). The F_1 hybrid and the later generation hybrid or backcross individual both had intermediate morphological scores along PC 1 (−0.254 and 0.440, respectively, Fig. 3).

The two fish with hybrid–backcross ITS 1 and GH RFLDs and intermediate morphology displayed the bull trout *Hind*III mtDNA RFLD (e.g., compare lanes 4 and 5 in Fig. 4 with lanes 5 and 6 in Fig. 5 and with lanes 3 and 4 in Fig. 6).

Discussion

Our morphological and molecular analyses of char from the Thutade Lake tributaries demonstrate that both Dolly Varden and bull trout are present within the watershed, and that the species hybridize in this undisturbed watershed. The discovery of Dolly Varden in the Thutade Lake watershed extends the natural range of Dolly Varden into the headwaters of the Peace River (MacKenzie system). The only other area where Dolly Varden are known from the Mackenzie system is in the upper Liard drainage (Haas and McPhail 1991). The early records (e.g., Lindsey 1956; Scott and Crossman 1973) of Dolly Varden in this system predate the recognition of bull trout as a distinct species and, in fact, all refer to bull trout. Dolly Varden probably entered the Thutade Lake watershed from the Skeena River system. The height of land between the Thutade Lake (Mackenzie Basin) watershed and the Sustut River in the Skeena system (Pacific Basin) is relatively low, and the presence of kokanee (*O. nerka*) in Thutade Lake implies a postglacial faunal transfer between these drainages.

When he resurrected the bull trout as a species separate from Dolly Varden, Cavender (1978) suggested that where their ranges overlap the two species may hybridize. This suggestion was based on two morphologically intermediate fish collected from the upper Skeena River (Swan and Morrison lakes). Since 1978, evidence has accumulated for hybridization among many species of *Salvelinus* (e.g., Leary et al. 1983; Hammer et al. 1989; Markle 1992; Wilson and Hebert 1993; Bernatchez et al. 1995). Thus, it is no surprise that Dolly Varden and bull trout hybridize; however, our data demonstrate that hybridization in char may extend beyond the F_1 generation (cf. Hammer et al. 1991). Further, our data suggest that morphological intermediacy alone is not necessarily reliable evidence of hybridization. For example, of the nine morphologically intermediate char from the Thutade Lake

© 1997 NRC Canada

Table 3. Summary of life history differences between sympatric Dolly Varden and bull trout in the Thutade Lake watershed (Bustard and Royea 1995; J.S. Baxter, personal observations).

Trait	Dolly Varden	Bull trout
Length at spawning (cm)	12–21 (mean = 15)	58–83 (mean = 72)
Spawning period	Early September to mid-October	Mid-August to early September
Watershed use	Resident in tributary streams	Migrate upstream from Thutade Lake to spawn (adfluvial)
Redd location	Upper watershed, groundwater seepage areas	Throughout watershed, areas of high cover
Redds per site	Grouped	Single
Redd area (m^2)	0.1	1.6
Water velocity at redds (cm s^{-1})	1–16	38–40
Water depth at redds (cm)	5–10	27–30
Redd gravel size (cm)	<2	2–4
Spawner distribution	Groups of pairs	Single pairs

watershed that we examined for molecular variation, only two showed rDNA and GH evidence of hybridization. This suggests that, in areas of sympatry, morphological differences that are reliable in allopatry must be interpreted with caution, and independent analyses (based on biparentally inherited molecular markers) are necessary to identify hybrids. This is especially true when introgression is occurring because such populations usually produce a mosaic of morphological types, and hybrids and backcrosses may resemble either or both of the parental species (Fig. 3; Neff and Smith 1979; Campton 1987).

Our data indicate that for the post-F_1 hybrid char the intensity of the Dolly Varden rDNA RFLD is much greater than that of the bull trout fragments (Fig. 5). The nondispersed organization of Dolly Varden and bull trout rDNA arrays (Phillips et al. 1989) means that the repeating unit is inherited essentially as a single locus. The differential intensity of the species-specific fragments could, however, result from crossing over within the rDNA repeat array to increase the proportion of one species' rDNA in some gametes of hybrids. The combination of morphological, rDNA, and GH RFLD analyses indicates that hybridization and probably at least some introgression is occurring between Dolly Varden and bull trout in the Thutade Lake watershed. For instance, the post-F_1-generation status of one of the hybrid individuals was confirmed by the GH RFLD analysis; it was homozygous for the bull trout allele at GH1, but heterozygous for Dolly Varden and bull trout alleles at GH2 (Fig. 6). This GH genotype could only result from a mating either between F_1 hybrids or between an F_1 or later generation hybrid and a bull trout (i.e., a backcross). Given the relative rarity of hybrids in the Thutade Lake watershed, it is more likely that an F_1 hybrid would backcross with one of the more abundant parental species than mate with another hybrid. Further, given equal numbers of parental genotypes and no gametic selection, the char that was homozygous for bull trout GH1 alleles and heterozygous for Dolly Varden and bull trout GH2 alleles is twice as likely to have resulted from a hybrid × bull trout backcross than from a hybrid × hybrid mating. The potential backcross identity of this hybrid char is consistent with its morphological score on PC 1 (0.440), which was displaced towards the bull trout end of the distribution (Fig. 3).

The char identifed as hybrids both exhibited the bull trout mtDNA *Hin*dIII restriction fragment pattern (Figs. 4, 5, and 6). Given the maternal inheritance of mtDNA, this implies matings between male Dolly Varden and female bull trout (for the F_1 hybrid) or, for the later generation hybrid or backcross individual, between male bull trout and female hybrids or between male and female hybrids. Although our ongoing sampling may reveal hybridization in both directions, what is known of the biology of the two species in the Thutade Lake watershed is consistent with asymmetrical hybridization being responsible for the F_1 hybrid. For instance, although mature bull trout are rare in the upper watershed (where immature and mature Dolly Varden predominate, Table 3), some bull trout do spawn there (Bustard and Royea 1995). Bull trout males are aggressive during spawning (McPhail and Murray 1979; Leggett 1980; Sexauer 1994; J.S. Baxter, personal observations). Usually, small males lose these aggressive encounters (McPhail and Murray 1979), and in the Thutade Lake watershed, Dolly Varden males are much smaller than mature bull trout (Table 3). In a variety of salmonids, including Dolly Varden and bull trout (McPhail and Murray 1979; Gross 1984, 1985; Hutchings and Myers 1985; Maekawa et al. 1993; Kitano et al. 1994; Sexauer 1994), small males resort to "sneaking" behaviour as a way of obtaining fertilizations. Our data suggest that hybrids in the Thutade Lake watershed could result from small Dolly Varden males sneaking fertilizations during bull trout spawnings. Similar sneaking mechanisms have been suggested to account for interspecific hybridization in other salmonids (e.g., McGowan and Davidson 1992; Kitano et al. 1994).

McPhail and Taylor (1995) found that adult hybrids between the species in the upper Skagit River (southwestern British Columbia) were similar in size to bull trout in that system, both of which are much larger than mature Dolly Varden. A similar sneaking mechanism, therefore, may account for asymmetry in hybrid or backcross matings as well (i.e., female hybrids × male Dolly Varden). Alternatively, if indeed hybrid char do tend to be large and thus more similar in size to bull trout, this could promote size-assortative matings between hybrids or between hybrids and bull trout. Such matings would account for post-F_1 hybrids bearing bull trout

© 1997 NRC Canada

mtDNA and recombinant GH genotypes that are homozygous for bull trout alleles.

Although our data do not permit us to evaluate the extent of hybridization and introgression between Dolly Varden and bull trout in the Thutade Lake watershed, the more extensive data of McPhail and Taylor (1995) from the upper Skagit River (southwestern British Columbia) indicate about 10% hybridization between these species. In the Thutade Lake system, hybridization does not appear to reach this level, but the F_1 hybrid and the backcross were found in two separate tributaries (South Pass and North Kemess creeks) and this suggests more than just a chance incident. In combination, the Thutade and Skagit data suggest that some hybridization and introgression probably occurs wherever the two species come into contact. Because all of the known areas of sympatry between Dolly Varden and bull trout are in recently glaciated areas, reproductive isolating mechanisms that prevent hybridization may still be evolving.

Notwithstanding our evidence of hybridization and introgression between Dolly Varden and bull trout, the species appear to be maintaining themselves as distinct genomes in the Thutade Lake watershed; they remain largely identifiable in ecological, morphological, and molecular genetic traits. Such differentiation in the face of gene flow implies that the hybrids are at some reproductive or ecological disadvantage relative to the parental species. The two species, therefore, may not necessarily completely introgress if they are in some form of gene flow-selection balance in the Thutade Lake watershed. The ecological differences between Dolly Varden and bull trout in the Thutade Lake watershed (Table 3) probably reflect polygenic genetic differences and hybrids would probably be ecologically intermediate. Such intermediacy of hybrids between Dolly Varden and bull trout may reduce their fitness relative to the parental species and stabilize introgression at some level or limit it to specific habitat patches (Arnold and Hedges 1995). Indeed, their ability to maintain themselves as distinct gene pools in the face of hybridization and some gene flow is the strongest evidence yet that Dolly Varden and bull trout are separate species.

Acknowledgements

We thank D. Bustard and R. Dams (David Bustard and Associates), D. Royea (Hallam Knight Piésold Ltd.), and El Condor Resources Ltd. for permission to study char in the Thutade Lake watershed. We are also grateful to the B.C. Ministry of Environment, Lands, and Parks (Fisheries Branch, Prince George) for permission to collect samples from the Thutade Lake area. We appreciate the comments on earlier versions of this paper by N.E. Down, G.R. Haas, J.R. Roome, C.E. Thompson, G.R. Smith, E.J. Crossman, and F.M. Utter. We are especially grateful to G.R. Haas for the tissues from laboratory-reared hybrids between Dolly Varden and bull trout, to D. O'Brien, K. Kostow, M. Rosenau, G. Thorpe, and M. Potyrala for providing tissue samples from outside Thutade Lake, and to C. Biagi for assistance in the laboratory. The research was funded, in part, by grants from the B.C. Ministry of Environment, Lands, and Parks (Research Section) and the UBC Fisheries Centre (to J.S.B.) and from the Natural Sciences and Engineering Research Council of Canada (to E.B.T. and J.D.M.).

References

Arnold, M.L., and Hedges, S.A. 1995. Are natural hybrids fit or unfit relative to their parents? Trends Ecol. Evol. **10**: 67–71.

Avise, J.C., and Lansman, R.A. 1983. Polymorphism of mitochondrial DNA in populations of higher animals. *In* Evolution of genes and proteins. *Edited by* M. Nei and R.K. Koehn. Sinauer Associates Incorporated, Sunderland, Mass. pp. 147–164.

Baker, R.J., Davis, S.K., Bradley, R.D., Hamilton, M.J., and Van den Bussche, R.A. 1989. Ribosomal-DNA, mitochondrial-DNA, chromosomal, and allozymic studies of a contract zone in the pocket gopher, *Geomys*. Evolution, **43**: 63–75.

Bernatchez, L., Glemet, H., Wilson, C.C., and Danzmann, R.G. 1995. Introgression and fixation of Arctic char (*Salvelinus alpinus*) mitochondrial genome in an allopatric population of brook trout (*Salvelinus fontinalis*). Can. J. Fish. Aquat. Sci. **52**: 179–185.

Brown, L.G. 1992. On the zoogeography and life history of Washington's native charr; Dolly Varden, *Salvelinus malma* (Walbaum) and bull trout, *Salvelinus confluentus* (Suckley). Appendix A. Bull trout/Dolly Varden management and recovery plan. Washington Department of Wildlife, Olympia, Wash.

Bustard, D., and Royea, D. 1995. Kemess South Gold–Copper Project 1994 Fisheries Studies. Report of Dave Bustard and Associates and Hallam Knight Piésold Limited to El Condor Resources Limited and St. Phillips Resources Incorporated, Vancouver, B.C.

Campton, D.E. 1987. Natural hybridization and introgression in fishes: methods of detection and genetic interpretations. *In* Population genetics and fishery management. *Edited by* N. Ryman and F.M. Utter. University of Washington Press, Seattle, Wash. pp. 161–192.

Cavender, T.M. 1978. Taxonomy and distribution of the bull trout, *Salvelinus confluentus* (Suckley), from the American Northwest. Calif. Fish Game, **64**: 139–174.

Cavender, T.M. 1984. Cytotaxonomy of North American *Salvelinus*. *In* Biology of the Arctic Charr: Proceedings of an International Symposium on Arctic Charr. *Edited by* L. Johnson and B.L. Burns. University of Manitoba Press, Winnipeg, Man. pp. 431–445.

Devlin, R.H. 1993. Sequence of sockeye salmon type 1 and 2 growth hormone genes and the relationship of rainbow trout with Atlantic and Pacific salmon. Can. J. Fish. Aquat. Sci. **50**: 1738–1748.

Feinberg, A.P., and Vogelstein, B. 1983. A technique for radiolabelling DNA restriction endonuclease fragments to high specific activity. Anal. Biochem. **132**: 6–13.

Grewe, P.M., Billington, N., and Hebert, P.D.N. 1990. Phylogenetic relationships among members of *Salvelinus* inferred from mitochondrial DNA divergence. Can. J. Fish. Aquat. Sci. **47**: 984–991.

Gross, M.R. 1984. Sunfish, salmon and the evolution of alternative reproductive strategies and tactics in fish. *In* Fish reproduction: strategies and tactics. *Edited by* G.W. Potts and R.J. Wootton. Academic Press, London. pp. 55–75.

Gross, M.R. 1985. Disruptive selection for alternative life histories in salmon. Nature (London), **313**: 47–48.

Haas, G.R., and McPhail, J.D. 1991. The systematics, zoogeography and evolution of Dolly Varden (*Salvelinus malma*) and bull trout (*Salvelinus confluentus*) in North America. Can. J. Fish. Aquat. Sci. **48**: 2191–2211.

Hammer, J., Dempson, J.B., and Skold, E. 1989. Natural hybridization between Arctic char (*Salvelinus alpinus*) and lake char (*S. namaycush*): evidence from northern Labrador. Nord. J. Freshwater Res. **65**: 54–70.

Hammer, J., Dempson, J.B., and Verspoor, E. 1991. Natural hybridization between Arctic char (*Salvelinus alpinus*) and brook trout

© 1997 NRC Canada

(*S. fontinalis*): evidence from northern Labrador. Can. J. Fish. Aquat. Sci. **48**: 1437–1445.

Herke, S.W., Kornfield, I., Moran, P., and Moring, J.R. 1990. Molecular confirmation of hybridization between northern pike (*Esox lucius*) and chain pickeral (*E. niger*). Copeia, **1990**: 846–850.

Hillis, D.M., and Dixon, M.T. 1991. Ribosomal DNA: molecular evolution and phylogenetic reconstruction. Q. Rev. Biol. **66**: 411–452.

Hutchings, J.A., and Myers, R.A. 1985. Mating between anadromous and nonanadromous Atlantic salmon, *Salmo salar*. Can. J. Zool. **63**: 2219–2221.

Kitano, S., Maekawa, K., Nakano, S., and Fausch, K.D. 1994. Spawning behaviour of bull trout in the upper Flathead drainage, Montana, with special reference to hybridization with brook trout. Trans. Am. Fish. Soc. **123**: 988–992.

Leary, R.F., Allendorf, F.W., and Knudsen, K.L. 1983. Consistently high meristic counts in natural hybrids between brook trout and bull trout. Syst. Zool. **32**: 369–376.

Leggett, J.W. 1980. Reproductive ecology and behaviour of Dolly Varden charr in British Columbia. *In* Charrs: salmonid fishes of the genus *Salvelinus*. *Edited by* E.K. Balon. W. Junk, The Hague, The Netherlands. pp. 721–737.

Lindsey, C.C. 1956. Distribution and taxonomy of fishes of the Mackenzie drainage of British Columbia. J. Fish. Res. Board Can. **13**: 759–789.

Maekawa, K., Hino, T., Nakano, S., and Smoker, W.W. 1993. Mate preference in anadromous and landlocked Dolly Varden (*Salvelinus malma*) females in two Alaskan streams. Can. J. Fish. Aquat. Sci. **50**: 2375–2379.

Markle, D.F. 1992. Evidence of bull trout × brook trout hybrids in Oregon. *In* Proceedings of the Gerhart Mountain Bull Trout Workshop. *Edited by* P.J. Howell and D.V. Buchanan. Oregon Chapter of the American Fisheries Society, Corvalis, Ore. pp. 58–67.

McGowan, C., and Davidson, W.S. 1992. Unidirectional natural hybridization between brown trout (*Salmo trutta*) and Atlantic salmon (*S. salar*) in Newfoundland. Can. J. Fish. Aquat. Sci. **49**: 1953-1958.

McPhail, J.D. 1961. A systematic study of the *Salvelinus alpinus* complex in North America. J. Fish. Res. Board Can. **18**: 793–816.

McPhail, J.D., and Murray, C.B. 1979. The early life history and ecology of Dolly Varden (*Salvelinus malma*) in the upper Arrow Lakes. Report of the Institute of Animal Resource Ecology and Department of Zoology (University of British Columbia) to BC Hydro and Ministry of Environment, Victoria, B.C.

McPhail, J.D., and Taylor, E.B. 1995. The Skagit char project. Report from the Department of Zoology, University of British Columbia, to the Skagit Environmental Endowment Commission, Seattle, Wash.

Morton, W.M. 1970. On the validity of all subspecific designations of North American *Salvelinus malma* (Walbaum). Copeia, **1970**: 581–587.

Neff, N.A., and Smith, G.R. 1979. Multivariate analysis of hybrid fishes. Syst. Zool. **28**: 176–196.

Phillips, R.B., Pleyte, K.A., and Ihssen, P.E. 1989. Patterns of chromosomal nucleolar organizer region (NOR) variation in fishes of the genus *Salvelinus*. Copeia, **1989**: 47–53.

Phillips, R.B., Manley, S.A., and Daniels, T.J. 1994. Systematics of the salmonid genus *Salvelinus* inferred from ribosomal DNA sequences. Can. J. Fish. Aquat. Sci. **51**(Suppl. 1): 198–204.

Pimentel, R.A. 1979. Morphometrics. The multivariate analysis of biological data. Kendall/Hunt Publishing Co., Dubuque, Iowa.

Pleyte, K., Duncan, S.D., and Phillips, R.B. 1992. Evolutionary relationships of the genus *Salvelinus* inferred from the DNA sequences of the first internal transcribed spacer (ITS 1) of ribosomal DNA. Mol. Phylogenet. Evol. **1**: 223–230.

Scott, W.B., and Crossman, E.J. 1973. Freshwater fishes of Canada. Bull. Fish. Res. Board Can. No. 184.

Sexauer, H.A. 1994. Life history aspects of bull trout, *Salvelinus confluentus*, in the eastern Cascades, Washington. M.Sc thesis, Central Washington University, Ellensburg, Wash.

Taylor, E.B., Foote, C.J., and Wood, C.C. 1996. Molecular genetic evidence for parallel life history evolution in a Pacific salmon (sockeye salmon and kokanee, *Oncorhynchus nerka*). Evolution, **50**: 401–416.

Wilson, C.C., and Hebert, P.D.N. 1993. Natural hybridization between Arctic char (*Salvelinus alpinus*) and lake trout (*S. namaycush*) in the Canadian Arctic. Can. J. Fish. Aquat. Sci. **50**: 2652–2658.

© 1997 NRC Canada

Does presmolt growth rate in steelhead trout (*Oncorhynchus mykiss*) and coho salmon (*Oncorhynchus kisutch*) predict growth rate in seawater?

Jörgen I. Johnsson, John Blackburn, W. Craig Clarke, and Ruth E. Withler

Abstract: The dependence of growth rate in postsmolts in seawater on presmolt growth rate was examined in steelhead trout (*Oncorhynchus mykiss*) and coho salmon (*Oncorhynchus kisutch*). Presmolt growth rate explained a significant but small proportion, 16% for steelhead and 6% for coho, of the variation in growth rate in seawater. The relationship between presmolt and postsmolt growth rate was positive for steelhead and slightly negative for coho. In steelhead trout, the relationship between freshwater and seawater growth rate was not affected by male parental type (steelhead or domesticated rainbow trout). In coho salmon, however, the slope of regression was affected by differences among family groups. These results demonstrate variable relationships between presmolt and postsmolt growth rate among salmonid species and families, suggesting that fast presmolt growth is not necessarily associated with fast seawater growth in hatchery-reared salmonids. These findings are of interest for rearing procedures in smolt hatcheries and for models predicting optimal time and size for migration in wild smolts.

Résumé : La relation de dépendance du taux de croissance après smoltification en mer sur le taux de croissance avant smoltification a été examinée chez la truite arc-en-ciel (*Oncorhynchus mykiss*) et le saumon coho (*Oncorhynchus kisutch*) . Le taux de croissance avant smoltification expliquait une partie statistiquement significative, mais tout de même faible (16% chez la truite arc-en-ciel et 6% chez le coho) de la variation du taux de croissance en mer. La relation entre les taux de croissance avant et après smoltification était positive chez la truite arc-en-ciel et légèrement négative chez le coho. Chez la truite arc-en-ciel, la relation entre le taux de croissance en eau douce et en eau salée ne subissait pas l'influence du type parental mâle (truite arc-en-ciel sauvage ou d'élevage). Toutefois, chez le saumon coho, la pente de régression variait en fonction des différences entre les groupes familiaux. Ces résultats mettent en évidence des différences du taux de croissance avant et après smoltification chez les différentes espèces et familles de salmonidés, et indiquent qu'une croissance rapide avant smoltification ne s'accompagne pas nécessairement d'une croissance rapide en mer chez les salmonidés élevés en écloserie. Ces constatations sont intéressantes du point de vue des méthodes d'élevage en écloserie et des modèles destinés à prévoir le moment et la taille optimales pour la migration des smolts sauvages.
[Traduit par la Rédaction]

Introduction

In anadromous salmonids, the relationship between presmolt growth rate and subsequent growth rate in seawater is of interest for (*i*) rearing procedures in smolt hatcheries, where it is important to know how selection for fast growth rate in the hatchery will affect adult sizes at return, and (*ii*) models predicting optimal time and size for migration in wild smolts (Bilton et al. 1982; Irwine and Ward 1989; Bohlin et al. 1993).

A model by Bohlin et al. (1993) suggests that the degree of dependence of postsmolt growth on presmolt growth is critical for the timing of the smolt migration. However, in spite of its importance, information on the relationship between presmolt and postsmolt growth is scarce. The available information on body size and growth for anadromous salmonids indicates a positive relationship between size at migration/release and adult size as well as between presmolt and postsmolt length increase (Elliot 1985; Berg and Berg 1989; Berg and Jonsson 1989; L'Abee-Lund et al. 1989; Hershberger et al. 1990). Nicieza and Braña (1993), however, found an inverse correlation between smolt size and marine length increment in Atlantic salmon (*Salmo salar*). Further, as pointed out by Bohlin et al. (1993), body sizes and body size increments can be positively correlated even if the corresponding specific growth rates are independent.

In two experiments with steelhead trout (*Oncorhynchus mykiss*) and coho salmon (*Oncorhynchus kisutch*) (Johnsson et al. 1993; R.E. Withler, unpublished data), which were designed for other purposes, we generated data on presmolt growth rates and growth rates after transfer to seawater. These data are used here to investigate the degree of correspondence between presmolt growth rates and postsmolt growth rates in seawater in these salmonids.

Received December 6, 1995. Accepted July 16, 1996.
J13195

J.I. Johnsson,[1] J. Blackburn, W.C. Clarke, and R.E. Withler. Department of Fisheries and Oceans, Biological Sciences Branch, Pacific Biological Station, Nanaimo, BC V9R 5K6, Canada.

[1] Present address: Section of Animal Ecology, Department of Zoology, Göteborg University, Medicinaregatan 18, S-413 90 Göteborg, Sweden.

© 1997 NRC Canada

Table 1. Summary of an ANCOVA on specific growth rate: (*a*) growth rate in seawater tanks in steelhead trout and steelhead/domesticated rainbow trout hybrids; male type is steelhead trout or rainbow trout; (*b*) growth rate in seawater net pens in coho salmon.

ANCOVA source	df	r^2 (%)	*F*	*P*
(*a*) Steelhead trout				
Male type	1	8.0	2.6	0.11
Freshwater growth	1	16.0	55.0	0.0001
Male type × freshwater growth	1	0.0	0.6	0.45
Replicate (male type)	8	8.7	4.2	0.0001
(*b*) Coho salmon				
Family	6	35.0	5.2	0.0001
Presmolt growth	1	6.3	22.4	0.0001
Family × presmolt growth	6	3.9	4.5	0.0002
Replicate (family)	7	1.0	1.0	0.45

Methods

Both experiments were conducted at the Pacific Biological Station in Nanaimo, B.C.

Steelhead experiment

In the laboratory, we monitored the growth of 18 maternal half-sib families of wild-type steelhead trout and steelhead/domesticated rainbow trout hybrids. The trout were reared under ambient temperature and simulated natural photoperiod conditions from June 1989 (first feeding) to late May 1990. On 28–29 August, all fish were tagged with passive integrated transponder tags (Johnsson et al. 1993). Throughout the experiments, the fish were fed to satiation using commercial salmon diet. On 8 December, 270 fish were distributed into ten 197-L fibreglass tanks. Five tanks contained steelhead trout (27 in each tank) and five contained hybrid trout (27 in each tank). On 23 April, when subsamples from the tanks demonstrated hypoosmoregulatory capacity in a seawater challenge test (Johnsson et al. 1994), all tanks were switched from freshwater to ambient 30 ppt seawater. The salinity change was completed in 30 min. Thereafter, all experimental fish were reared in seawater until the termination of the experiment on 24 May 1990.

During the experiments, each individual was weighed at regular intervals. Growth was divided into two periods: growth in freshwater from 5 March to 15 April (mean temperature (T) = 6.9°C) and growth in seawater from 15 April to the termination of the experiment on 24 May (T = 9.4°C). On 5 March, mean masses were 55.4 (±1.6 SE) g for steelhead trout and 99.4 (±3.1 SE) g for hybrid trout, respectively. Final mean masses were 107.7 (±2.9 SE) g for steelhead trout and 178.1 (±5.9 SE) g for hybrid trout, respectively. For further details, see Johnsson et al. (1993).

Coho experiment

On 20 January, coho fry from a third generation selected Kitimat River stock were ponded into 200-L tanks. Feed was supplied continuously during daylight hours by automatic feeders. The photoperiod was maintained at 10 h from 20 January to 23 March when it was increased to 13.5 h and thereafter adjusted to simulate a seasonally increasing natural daylight plus civil twilight in order to produce underyearling smolts (Clarke 1992). On 30 March, 404 fry were PIT tagged. The fish were gradually acclimated to seawater according to the following schedule: 10 ppt on 21 March, 15 ppt on 7 April, 20 ppt on 2 May, and 29 ppt on 16 May. From 28 January to 18 May, temperature was 14°C. The fish were moved out to two replicate seawater netpens on 20 May. The temperature in the seapens was 14°C on 20 May, rising to 18°C in July and then decreasing to 13.5°C on 4 October; the mean for the entire period was 15.8°C. Wet mass was measured for each individual on 6 April, 10–11 May, and 3–5 October. The mean mass (replicates pooled) was 8.5 (±0.1 SE) g on 6 April, 24.5 (±0.3 SE) g on 10 May, and 362.1 (±4.3 SE) g on 4 October.

Growth calculations and statistical analyses

In both experiments, growth data was subjected to GLM analysis of covariance (ANCOVA) (SAS Institute Inc. 1985). Specific growth rates (Ricker 1979) were calculated as $G = \ln(M_{t1}/M_{t0}) \times 100d^{-1}$ where M_{t0} and M_{t1} were the initial and final wet mass for the period and d was the time in days.

Results

Steelhead experiment

About a third of the total variation in seawater growth of steelhead trout was explained by the ANCOVA model (Table 1*a*). Seawater growth rate (G_s) was significantly correlated with preceding freshwater growth rate (G_f) (r^2 = 16%), and the estimated regression equation was G_s = 0.6(±0.1 SE) + 0.4(±0.1 SE)G_f. There was no significant effect of male parental type (steelhead or domesticated rainbow trout) or male parent by freshwater growth rate interaction. However, there was a significant variation among replicate tanks within male type.

Coho experiment

Almost half of the total variation in seawater growth of coho salmon was explained by the model (Table 1b). Seawater growth rate (G_s) was significantly predicted by presmolt growth in brackish water (G_b) (r^2 = 6%), but the effect of family explained more of the growth variation (r^2 = 35.0%). A significant interaction between family and presmolt growth indicated that the slope between presmolt growth and seawater growth was heterogenous among families. The regression slope was negative in six families and positive in one. The overall regression equation was G_s = 2.4(±0.2 SE) – 0.1(±0.05 SE)G_b.

© 1997 NRC Canada

Discussion

Our results demonstrate that postsmolt seawater growth rate in steelhead trout and coho salmon is significantly predicted by presmolt growth rate in freshwater. However, it is interesting to note the following. (*i*) The dependence of seawater growth on freshwater growth was not very strong ($b \sim 0.4$ for steelhead and $b \sim -0.1$ for coho, respectively). (*ii*) Only a small proportion of the total variation in seawater growth was explained by preceding presmolt growth (16% for steelhead and 6% for coho, respectively). This indicates that although presmolt growth rate is correlated significantly with subsequent growth in seawater, the relationship is weakened by a large amount of random variation, limiting the use of freshwater growth rate as an indicator of future growth performance in the sea. (*iii*) The relationship between presmolt and postsmolt growth rate differed between experiments, being positive in the steelhead experiment and slightly negative in the coho experiment. There was also considerable variation among family groups in the coho experiment whereas there was no effect of male parental life-history type (steelhead or domesticated rainbow trout) in the steelhead experiment. Overall, our results suggest that fast presmolt growth is not necessarily associated with fast seawater growth.

There are several possible explanations for the absence of consistent and strong positive correlations between freshwater and seawater growth rates in the present study. Firstly, growth-promoting characters may be specific to a particular environment. For example, juvenile coho salmon rearing pelagically in lakes are less aggressive than nearby stream-living coho (Swain and Holtby 1989), and chinook salmon (*Oncorhynchus tshawytscha*) populations with longer stream residence are more aggressive than ocean-type chinook (Taylor 1990). Thus, inherited (nonplastic) behavioural characters may have differential growth-promoting effects in stream and pelagic environments, respectively. Furthermore, the risk of predation in the food-limited stream environment may act to favour slower growth compared with the marine environment with its greater availability of food (Johnsson and Abrahams 1991). Similar limitations on phenotypic plasticity may constrain physiological adaptations, such as osmoregulation (Hasegawa et al. 1987) and digestion, from being equally effective in freshwater and seawater.

Secondly, in our experiments, as in most hatcheries, all fish were placed in seawater simultaneously, whereas in natural populations, migration is "voluntary" and individuals of different age and size migrate at different times (Bohlin et al. 1993). It is likely that the possibility of individual choice of migration time might influence the correlation between presmolt and postsmolt growth.

Thirdly, higher food abundance in the sea compared with the stream would elevate the intercept of the growth regression equation. In contrast with wild smolts that experience a limited food supply in streams, the fish in our experiments experienced no difference in food abundance between the freshwater and seawater environments.

Fourthly, some of the differences may be attributed to the different designs of the two experiments. In the steelhead experiment, seawater growth was measured over a shorter time (39 days) and may not be representative of long-term growth in seawater. The steelhead were also larger than coho when transferred to seawater. Finally, the steelhead were reared in a uniform tank enviroment during the entire experiment whereas coho were transferred from tanks to seawater netpens.

It would be interesting to compare our results with other studies on premigratory and postmigratory growth rates of hatchery and wild salmonid populations. Presently, this kind of information is lacking, providing an unexplored and important field of research.

Acknowledgements

This study was made possible by a grant (40.0357/93) to J.I.J. from the Swedish Council for Forestry and Agricultural Research. Excellent technical assistance was provided by the staff at the Pacific Biological Station in Nanaimo, B.C. We are grateful to Torgny Bohlin for stimulating discussions during this project.

References

Berg, O.K., and Berg, M. 1989. Sea growth and time of migration of anadromous Arctic char (*Salvelinus alpinus*) from the Vardnes River, in northern Norway. Can. J. Fish. Aquat. Sci. **46**: 955–960.

Berg, O.K., and Jonsson, B. 1989. Growth and survival rates of the anadromous trout, *Salmo trutta*, from the Vardnes River, northern Norway. Environ. Biol. Fishes, **29**: 145–154.

Bilton, H.T., Alderdice, D.F., and Schnute, J.T. 1982. Influence of time and size at release of juvenile coho salmon (*Oncorhynchus kisutch*) on returns at maturity. Can. J. Fish. Aquat. Sci. **39**: 426–447.

Bohlin, T., Dellefors, C., and Faremo, U. 1993. Optimal time and size for smolt migration in wild sea trout (*Salmo trutta*). Can. J. Fish. Aquat. Sci. **50**: 224–232.

Clarke, W.C. 1992. Environmental factors in the production of Pacific salmon smolts. World Aquacult. **23**(4): 40–42.

Elliot, J.M. 1985. Growth, size, biomass and production for different life-stages of migratory brown trout *Salmo trutta* in a lake district stream, 1966–83. J. Anim. Ecol. **54**: 985–1001.

Hasegawa, S., Hirano, T., Ogasawara, T. Iwata, M., Akiyama, T., and Arai., S. 1987. Osmoregulatory ability of chum salmon, *Oncorhynchus keta*, reared in freshwater for prolonged periods. Fish Physiol. Biochem. **4**: 101–110.

Hershberger, W.K., Myers, J.M., Iwamoto, R.N., Mcauley, W.C., and Saxton, A.M. 1990. Genetic changes in the growth of coho salmon (*Oncorhynchus kisutch*) in marine net pens, produced by ten years of selection. Aquaculture, **85**: 187–197.

Irwine, J.R., and Ward, B.R. 1989. Patterns of timing and size of wild coho salmon (*Oncorhynchus kisutch*) smolts migrating from the Keogh River Watershed on northern Vancouver Island. Can. J. Fish. Aquat. Sci. **46**: 1086–1094.

Johnsson, J.I., and Abrahams, M.V. 1991. Interbreeding with domestic strain increases foraging under threat of predation in juvenile steelhead trout (*Oncorhynchus mykiss*): an experimental study. Can. J. Fish. Aquat. Sci. **48**: 243–247.

Johnsson, J.I., Clarke, W.C., and Withler, R.E. 1993. Hybridization with domesticated rainbow trout (*Oncorhynchus mykiss*) reduces seasonal variation in growth of steelhead trout (*O. mykiss*). Can. J. Fish. Aquat. Sci. **50**: 480–487.

Johnsson, J.I., Clarke, W.C., and Blackburn, J. 1994. Hybridization with domesticated rainbow trout reduces seasonal variation in seawater adaptability of steelhead trout (*Oncorhynchus mykiss*). Aquaculture, **121**: 73–77.

L'Abee-Lund, J.H., Jonsson, B., Jensen, A.J., Sættem, L.M., Heggberget, T.G., Johnsen, B.O., and Næsje, T.F. 1989.

© 1997 NRC Canada

Latitudinal variation in life-history characteristics of sea-run migrant brown trout *Salmo trutta*. J. Anim. Ecol. **58**: 525–542.

Nicieza, A.G., and Braña, F. 1993. Relationships among smolt size, marine growth, and sea age at maturity of Atlantic salmon (*Salmo salar*) in northern Spain. Can. J. Fish. Aquat. Sci. **50**: 1632–1640.

Ricker, W.E. 1979. Growth rates and models. *In* Fish physiology. Vol. VIII. Bioenergetics and growth. *Edited by* W.S. Hoar, D.J. Randall, and J.R. Brett. Academic Press, New York. pp. 677–743.

SAS Institute Inc. 1985. SAS procedures guide for personal computers, version 6 edition. SAS Institute Inc., Cary, N.C.

Swain, D.P., and Holtby, L.B. 1989. Differences in morphology and behavior between juvenile coho salmon (*Oncorhynchus kisutch*) rearing in a lake or in its tributary stream. Can. J. Fish. Aquat. Sci. **46**: 1406–1414.

Taylor, E.B. 1990. Variability in agonistic behaviour and salinity tolerance between and within two populations of juvenile chinook salmon, *Oncorhynchus tshawytscha*, with contrasting life histories. Can. J. Fish. Aquat. Sci. **47**: 2172–2180.

© 1997 NRC Canada

Production of labile and refractory dissolved organic carbon by zooplankton excretion: an experimental study using large outdoor continuous flow-through ponds

Je-Chul Park, M. Aizaki, T. Fukushima, and A. Otsuki

Abstract: Experiments using six outdoor experimental ponds equipped with a continuous flow-through system were conducted from 6 to 31 August 1992 to determine whether zooplankton grazing activity can be a significant source of labile (L-DOC) and refractory dissolved organic carbon (R-DOC) in a eutrophic shallow lake. During large variations of chlorophyll *a* concentration caused by active zooplankton grazing pressure, distinct increases in total dissolved organic carbon (T-DOC) concentrations were observed in all ponds. Subsequently, pond T-DOC concentrations fluctuated at levels higher than those in inflow water, which remained almost constant throughout the experimental period. The T-DOC fluctuations were closely related to changes in zooplankton density. The correlation between L-DOC production rates and zooplankton densities in the ponds was highly significant ($r = 0.74$, $P < 0.01$). Incubation experiments with filtrates of pond water showed that R-DOC, defined as T-DOC remaining after 100 days of incubation in the dark at 20°C, was produced during active zooplankton grazing. The proportion of R-DOC in T-DOC was almost constant, irrespective of the N:P loading ratios (5, 10, and 20 by weight) in inflow waters.

Résumé : Des expériences réalisées dans six bassins expérimentaux extérieurs munis de systèmes d'écoulement continu de l'eau ont été réalisées du 6 au 31 août 1992 pour déterminer si l'activité de broutage du zooplancton peut être une source importante de carbone organique dissous labile (COD-L) ou réfractaire (COD-R) dans un lac eutrophe peu profond. Au cours des grandes variations de la concentration de la chlorophylle *a* liées à la pression exercée par un broutage actif du zooplancton, on a observé une nette augmentation du carbone organique dissous total (COD-T) dans tous les bassins. Par la suite, la concentration du COD-T a fluctué à des concentrations supérieures à celle qui a été mesurée dans l'eau d'alimentation, concentration qui est demeurée presque constante pendant toute la période de l'étude. Les fluctuations de la COD-T étaient étroitement liées aux changements de densité du zooplancton. La corrélation entre le taux de production de COD-L et la densité du zooplancton dans les bassins était statistiquement hautement significative ($r = 0{,}74$, $P < 0{,}01$). Les expériences d'incubation avec des filtrats obtenus à partir de l'eau des bassins ont montré que le COD-R, défini comme étant la COD-T qui reste après 100 jours d'incubation à l'obscurité à 20°C, était produit durant le broutage actif par le zooplancton. La proportion de COD-R dans la COD-T était presque constante, quels qu'aient été les rapports des charges en N:P (5, 10 et 20, en poids) dans l'eau d'alimentation.
[Traduit par la Rédaction]

Introduction

The quality of drinking water in reservoirs and lakes is greatly affected by the concentration of dissolved organic matter because of the unpleasant taste and (or) odor caused by chlorination. At the same time, from a viewpoint of aquatic energy flow, the dissolved organic matter, especially the labile component, can be an important energy source for bacteria, which then flows to higher trophic levels in lake ecosystems (Wetzel 1992). It is important therefore to identify the sources and nature of dissolved organic matter in reservoirs and lakes.

Received December 21, 1995. Accepted July 15, 1996.
J13219

Je-Chul Park and A. Otsuki. Tokyo University of Fisheries, 4-5-7 Konan, Minato-ku, Tokyo 108, Japan.
M. Aizaki and T. Fukushima. National Institute for Environmental Studies, 16-2 Onogawa, Tsukuba, Ibaraki 305, Japan.

Dissolved organic carbon (DOC) in natural water is often categorized with respect to bacterial degradability into labile (L-DOC) and refractory DOC (R-DOC). Recently, many studies have been carried out to clarify the role of DOC in lake waters (Sondergaard 1984; Munster 1985; Schindler et al. 1992; Sondergaard and Borch 1992; Sondergaard et al. 1995), and the major portion of the DOC pool has been reported to be R-DOC (Wetzel 1983; Sondergaard 1984; Sondergaard and Borch 1992). In contrast, photosynthetically excreted DOC (E-DOC) has been shown to occupy only a minor portion of the DOC pool (Sondergaard et al. 1995) because of its rapid use by bacteria (Brock and Clyne 1984; Bratbak 1987; Sondergaard et al. 1988; Nixdorf 1990; Hobbie 1992; Middelboe et al. 1992).

There are two major sources of DOC in lake water: allochthonous DOC from catchment and autochthonous DOC produced within lakes. Several studies showed that the origin of R-DOC was allochthonous (Wetzel 1983; Munster and Chrost 1990; Meili 1992). However, the fact that the concentrations of R-DOC in eutrophic, shallow Lake Kasumigaura were

© 1997 NRC Canada

always higher than those in inflowing river waters suggested that part of this R-DOC was being produced autochthonously (Fukushima et al. 1996).

The main purpose of the present study was to experimentally evaluate in six outdoor ponds with a continuous flow-through system whether zooplankton grazing activity can be the partial source of L-DOC and R-DOC in eutrophic, shallow lakes where standing stocks of phytoplankton are sufficient to support zooplankton.

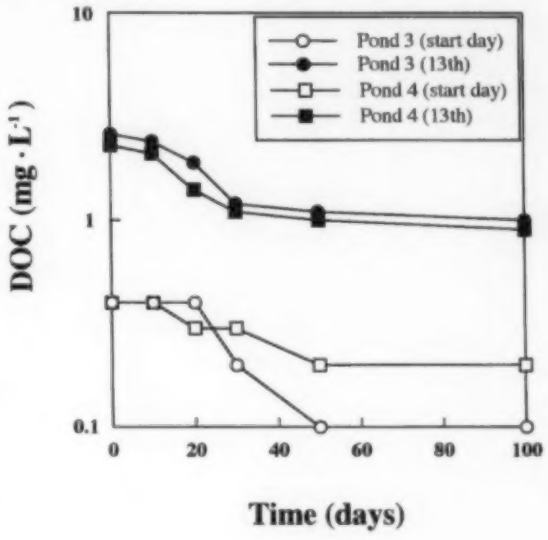
Materials and methods

The experiments were conducted in six outdoor experimental ponds (P1–P6) of regular hexagonal shape, 1.7 m in depth, and 36 m^3 in volume. These ponds were situated at about 0.5 km from Lake Kasumigaura's shore. Before the experiment, the ponds were lined with a PVC-covered nylon fabric and filled with sand-filtered water from Lake Kasumigaura. This sand-filtered lake water (inflow water) contained less than 1 μg chlorophyll $a \cdot L^{-1}$ and flowed continuously into the ponds. Flow rates were adjusted to renew the pond water every 20 days (1.25 $L \cdot min^{-1}$). The N:P loading ratios of inflow water were adjusted to 5 (P1 and P2), 10 (P3 and P4), and 20 (P5 and P6) by weight by the continuous addition of nutrient solutions with a peristaltic pump. The phosphorus concentration in the enrichment solution was fixed at 0.46 mg $P \cdot L^{-1}$ (KH_2PO_4) whereas three different concentrations (2.3, 4.6, and 9.2 mg $N \cdot L^{-1}$) of NH_4NO_3 were added to achieve the respective N:P loading ratios. Each N:P loading ratio was achieved in two ponds.

At the start of the experiment, about 20 L of unfiltered Lake Kasumigaura water containing mainly *Microcystis* sp. was inoculated into each pond. Zooplankton and water samples were taken at the center of each pond with a 1.2-m column water sampler from 6 to 31 August 1992. In P3, additional sampling was conducted at 4-h intervals from 7 to 8 August to confirm the release of DOC by photosynthetic activity of phytoplankton. Zooplankton was sampled three times a week at different intervals, and phytoplankton and water were sampled daily. The zooplankton samples were screened through a 100-μm mesh net and preserved with formaldehyde. We used the 100-μm mesh net to collect large-size zooplankton in pond waters because *Microcystis* colonies of 20–60 μm in diameter form about 80% of all colonies and would be suitable foods for large-size zooplankton (Okada and Aiba 1983; Hanazato and Yasuno 1987). Phytoplankton samples were fixed with formaldehyde. Zooplankton and phytoplankton species were identified and counted under a microscope. Water samples were filtered through a Whatman GF/F glass fiber filter (precombusted at 450°C for 3 h) immediately at the laboratory. A portion (200 mL) of the filtrate was poured into precombusted 250-mL glass bottles (500°C, 6 h) and incubated in the dark at 20°C for 100 days to determine R-DOC in the filtrate. Subsamples (10 mL) were collected from the 250-mL bottles at selected times, 0, 10, 20, 30, 50, and 100 days, and used for determination of DOC without filtration because the particulate organic carbon (POC) content (mainly bacterial carbon) was always less than 0.1 mg $C \cdot L^{-1}$. The difference between the initial T-DOC and R-DOC was defined as L-DOC (Ogura 1972; Satoh and Abe 1987; Servais et al. 1987, 1989).

Assuming that L-DOC and R-DOC in pond water were produced by separate biological activities, we calculated the production rates of L-DOC and R-DOC (PL and PR) by the following two equations to examine how they differed:

Fig. 1. Time versus logarithm of DOC during the biodegradation tests for P3 and P4.



$$(1) \quad dCL/dt = PL + Q/V(CiL - CL) - CLk_1$$

where V is the pond volume (cubic metres), Q is the inflow rate (cubic metres per day), t is time (day), PL is the L-DOC production rate (grams carbon per cubic metre per day), CL is the pond water L-DOC concentration (grams carbon per cubic metre), CiL is the inflow water L-DOC concentration (grams carbon per cubic metre), and k_1 is the decomposition rate constant for L-DOC and

$$(2) \quad dCR/dt = PR + Q/V(CiR - CR) - CRk_2$$

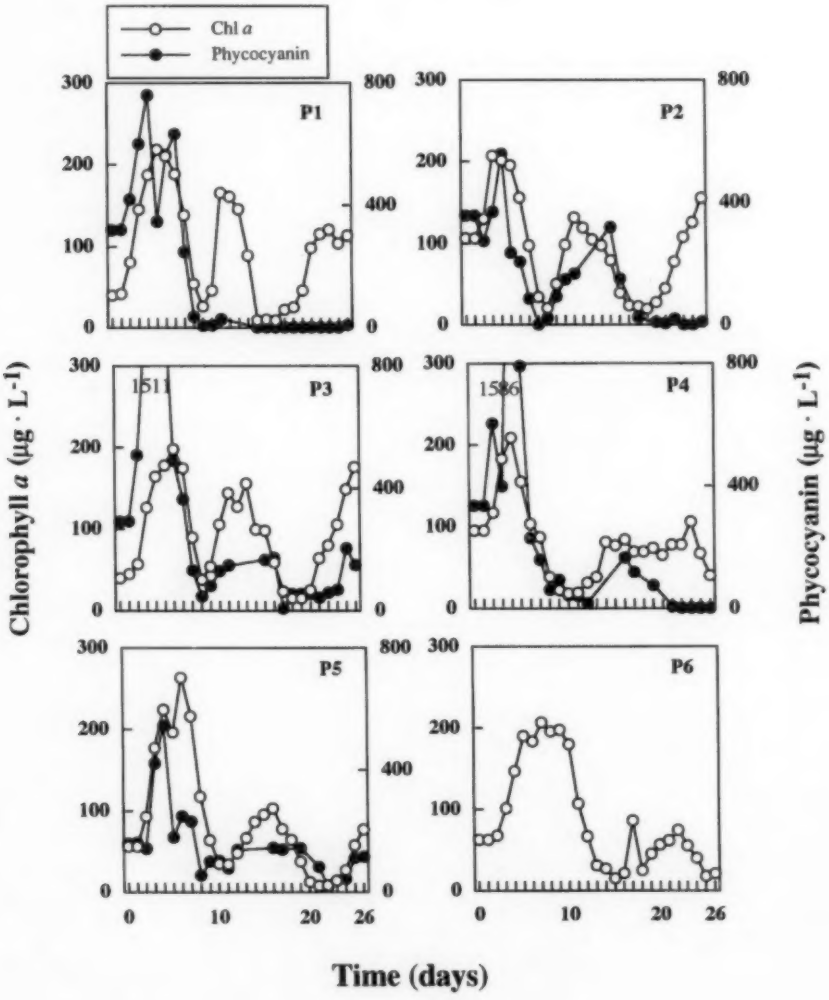
where CR and CiR are the R-DOC concentrations (grams carbon per cubic metre) in pond and inflow waters, respectively, PR is the R-DOC production rate (grams carbon per cubic metre per day), and k_2 is the decomposition rate constant for R-DOC. The production rate of T-DOC was estimated as the sum of eq. 1 and eq. 2.

The decomposition constants k_1 and k_2 were estimated from the slope of two linear regressions of T-DOC and time in logarithmic graphs, based on the decomposition experiments of T-DOC in pond water filtrates in the dark at 20°C for 100 days. In eq. 2, k_2 was assumed to be zero because of the extremely slow decomposition rate of R-DOC (Fig. 1).

DOC concentration was measured with a Shimadzu TOC 5000 high-temperature catalyst analyzer. The precision was within a range of ±0.05 mg $C \cdot L^{-1}$ for DOC concentrations less than 10 mg $C \cdot L^{-1}$. Concentrations of chlorophyll *a* and phycocyanin were estimated by extraction with a 100% methanol (Marker et al. 1980) and a 10 mM phosphate buffer solution (pH 7.0) by the HPLC-fluorometric method (Otsuki et al.

© 1997 NRC Canada

Fig. 2. Chlorophyll *a* and phycocyanin concentrations in ponds P1–P6.



1994), respectively. Dissolved inorganic phosphorus (DIP) (PO_4-P) and dissolved inorganic nitrogen (DIN) (NH_4-N, NO_3-N) were measured using a Technicon Auto-Analyzer AAII.

Results and discussion

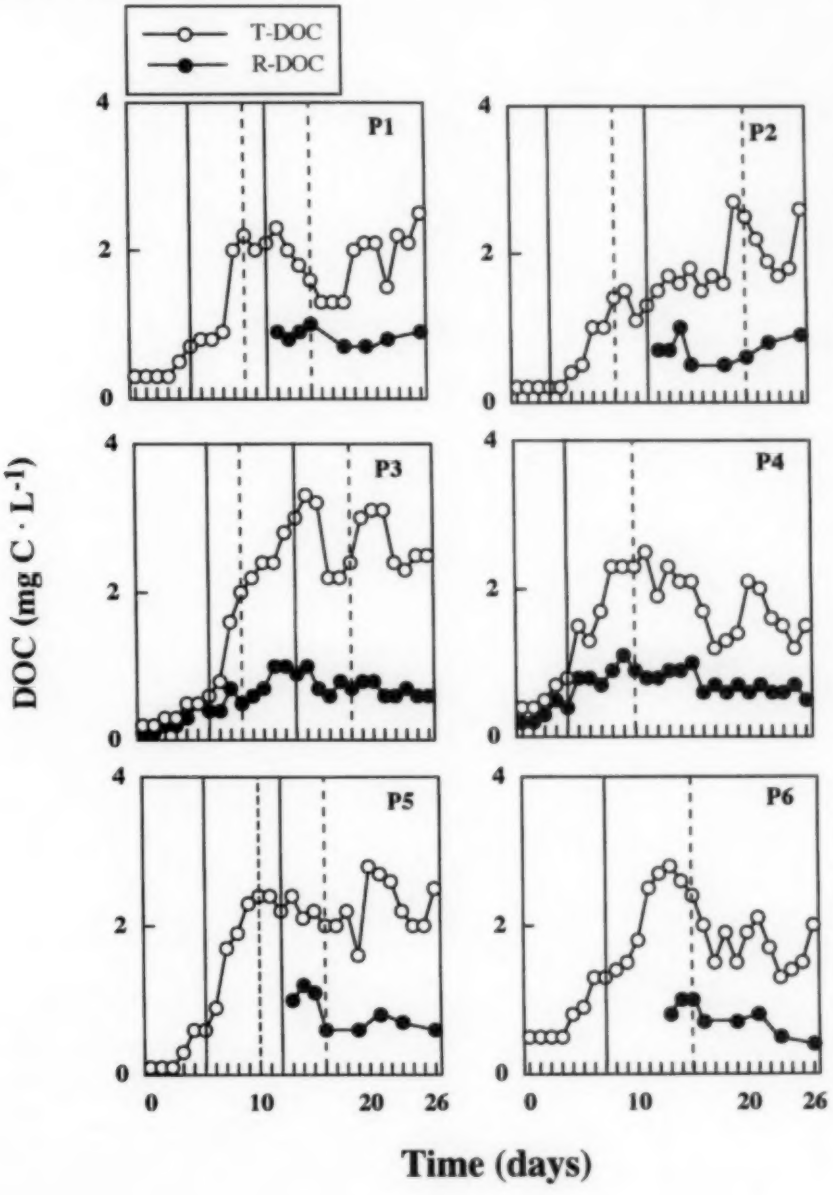
We tried to establish different levels of cyanobacterial blooms by continuously adding nutrient solutions with three different N:P ratios to set up different concentration of T-DOC (Fig. 1). However, different levels were not established. It seemed that the changes in the six ponds occurred irrespective of different N:P ratio loadings.

Changes in chlorophyll *a* and phycocyanin concentrations and phytoplankton species composition

The trends in chlorophyll *a* and phycocyanin concentrations in all ponds were similar (Fig. 2). During the initial 10 days of this experiment, the first peaks of chlorophyll *a* and phycocyanin were observed in all ponds, except for P6 in which phycocyanin was not measured. The peak concentrations of chlorophyll *a* and phycocyanin ranged from 198 to 263 $\mu g \cdot L^{-1}$ and from 558 to 1586 $mg \cdot L^{-1}$ in all ponds, respectively. The dominant phytoplankton genera during the first chlorophyll *a* peaks in all ponds (between the 2nd and 12th days) were cyanobacteria (mainly *Microcystis* sp. and *Oscillatoria* sp.), accounting for about 50% of total phytoplankton

© 1997 NRC Canada

Fig. 3. Daily T-DOC and R-DOC concentrations in ponds P1–P6. Solid vertical lines indicate the dates of chlorophyll *a* peaks and broken lines indicate the dates of chlorophyll *a* minima.



cell numbers (Table 1). Although a second chlorophyll *a* peak also appeared in all ponds between the 12th and 18th days, large peaks of phycocyanin were not observed, indicating changes in the dominant phytoplankton from cyanobacteria to the other genera. After the first and second peaks of chlorophyll *a*, the dominant phytoplankton clearly changed to green algae, *Coelastrum* sp., *Scenedesmus* sp., and *Kirchneriella* sp. in all ponds.

Changes in T-DOC and R-DOC concentrations

Although T-DOC concentrations (average 2.3 ± 0.2 mg $C \cdot L^{-1}$) in inflow water remained almost constant throughout the experimental period and R-DOC made up 82–95% of T-DOC (Table 2), T-DOC in all ponds gradually increased during the first 5 days of the experiment and then increased rapidly (Fig. 3). The maximum net increases in concentrations of T-DOC, calculated by subtracting T-DOC in inflowing

© 1997 NRC Canada

Table 1. Changes in dominant phytoplankton species on five dates in ponds P1–P6.

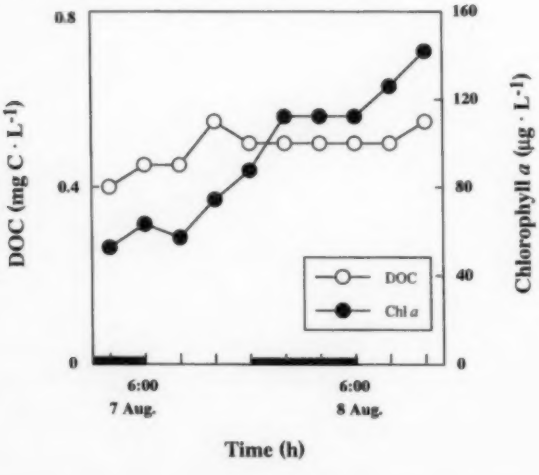
Date		P1	P2	P3	P4	P5	P6
6 Aug.	Total no. (mL^{-1})	1.4×10^5	2.8×10^5	8.4×10^4	1.7×10^5	1.5×10^5	1.1×10^5
	Dominant	Oscilla.	Micro.	Oscilla.	Oscilla.	Oscilla.	Micro.
	Percentage	55.5	59.8	54.0	29.4	47.6	49.2
12 Aug.	Total no. (mL^{-1})	2.5×10^5	6.9×10^4	2.2×10^5	9.6×10^4	1.3×10^5	2.0×10^5
	Dominant	Micro.	Micro.	Micro.	Coela.	Kirch.	Micro.
	Percentage	47.5	43.7	62.1	39.9	57.6	54.1
19 Aug.	Total no. (mL^{-1})	7.6×10^4	6.7×10^4	8.5×10^4	1.1×10^5	1.9×10^5	4.4×10^4
	Dominant	Kirch.	Micro.	Micro.	Phorum.	Coela.	Micro.
	Percentage	67.0	51.2	60.1	44.6	42.6	39.9
26 Aug.	Total no. (mL^{-1})	4.0×10^4	4.2×10^4	1.3×10^4	4.2×10^4	1.5×10^4	2.2×10^5
	Dominant	Coela.	Coela.	Pedia.	Scened.	Osilla.	Coela.
	Percentage	35.7	58.5	21.2	63.6	37.6	44.5
31 Aug.	Total no. (mL^{-1})	9.1×10^4	1.6×10^5	1.5×10^5	2.2×10^4	4.9×10^4	1.9×10^4
	Dominant	Coela.	Coela.	Kirch.	Kirch.	Kirch.	Scened.
	Percentage	77.6	66.4	61.2	61.3	25.2	33.3

Note: Micro., *Microcystis* sp.; Oscilla., *Oscillatoria* sp.; Phorum., *Phorumidium mucicola*; Kirch., *Kirchneriella* sp.; Coela., *Coelastrum* sp.; Scened., *Scenedesmus* sp.; Pedia., *Pediasturum duplex*.

Table 2. R-DOC as a percentage of T-DOC on the first day and at steady state (average during the 10th to 20th days).

	First day			Steady state		
Pond	T-DOC (mg $C \cdot L^{-1}$)	R-DOC (%)	Inflow DOC (%)	T-DOC (mg $C \cdot L^{-1}$)	R-DOC (%)	Inflow DOC (%)
P1	0.3	8	88	1.9	18	53
P2	0.2	8	88	1.8	17	56
P3	0.3	8	88	2.6	14	47
P4	0.4	7	85	1.9	19	55
P5	0.1	4	95	2.2	18	51
P6	0.5	10	82	2.0	19	53

Fig. 4. Diurnal changes in DOC and chlorophyll *a* concentrations during the initial period of the experiment in P3.



water from T-DOC in pond water, reached 3.3 and 2.7 mg $C \cdot L^{-1}$ in P3 and P6, respectively. After the increased T-DOC concentrations approached their maxima between the 10th and 15th days, they fluctuated at levels higher than that in inflow water, seemingly in an approximate steady state (Fig. 3). The increased R-DOC concentrations also remained largely unchanged after the 13th day in all ponds (Table 2).

The distinct increase in T-DOC concentration (both L-DOC and R-DOC increase) occurred simultaneously with rapid decreases in algal biomass. Although chlorophyll *a* concentration during the initial 5 days of the experiment increased rapidly (Fig. 2), T-DOC concentration scarcely changed. However, when the first phytoplankton bloom waned (chlorophyll *a* declined to a minimum) as the result of zooplankton grazing activity during the 8th to 10th days, rapid increases in T-DOC and R-DOC concentrations clearly occurred in all ponds, except for P6 in which the decrease in chlorophyll *a* concentration was delayed (Fig. 2). The net increase in L-DOC (L-DOC in inflowing water was subtracted from that in pond water) ranged from 0.9 to 1.8 mg $C \cdot L^{-1}$ and that in

© 1997 NRC Canada

Fig. 5. Daily change in DOC concentration in relation to daily change in chlorophyll *a* concentration in ponds P1–P6. Cyanobacteria, during the initial 10 days; green algae, during the 18th to 26th days.

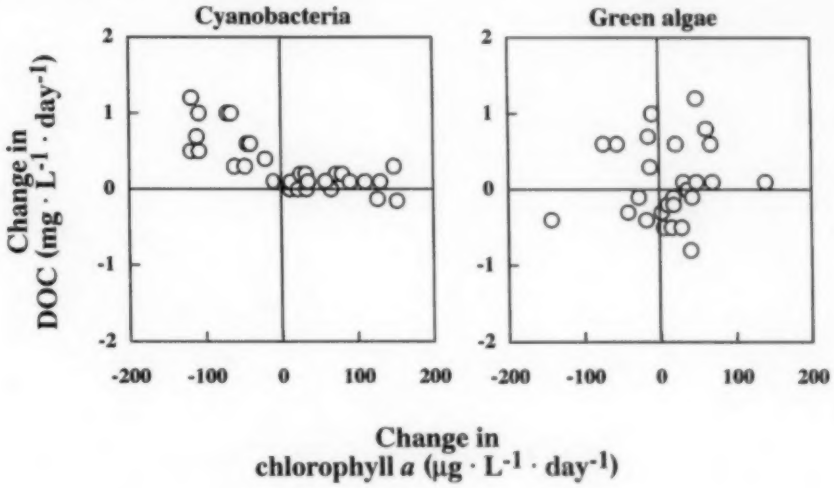
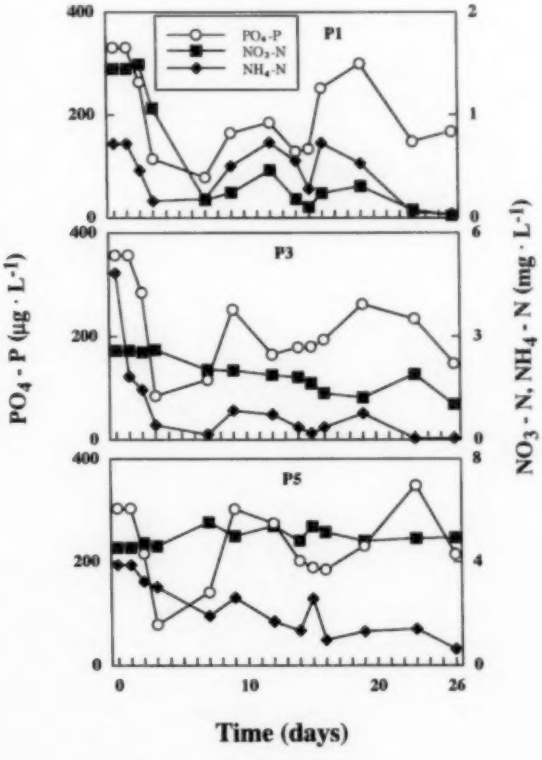


Fig. 6. Daily PO_4-P, NH_4-N, and NO_3-N concentrations in P1, P3, and P5.



R-DOC ranged from 0.7 to 1.0 mg C · L^{-1} (calculated similarly). Subsequently, minima in chlorophyll *a* concentrations roughly coincided with the peaks of the net increases in T-DOC in all ponds.

Although chlorophyll *a* concentration in P3 during the initial period of the experiment rapidly increased to about three times its initial value after only 36 h, the T-DOC concentration scarcely increased during this period (Fig. 4). This result suggests that E-DOC was quickly utilized by bacteria, as many researchers have previously pointed out (Brock and Clyne 1984; Bratbak 1987; Sondergaard et al. 1988; Nixdorf 1990; Hobbie 1992; Middelboe et al. 1992). That is, if the E-DOC production depends on the observed increase in chlorophyll *a* concentration, a much larger net increase in T-DOC concentration should have been observed in all ponds during the initial 2–4 days of the experiment. Thus, the L-DOC in the filtrates in the present study would not include E-DOC because of its extremely rapid mineralization by bacteria, although E-DOC has been reported to account for 0–70% of the total carbon fixed by phytoplankton (Chrost and Faust 1983; Watanabe 1984; Jensen et al. 1985; Chrost 1986; Hama and Handa 1987; Baines and Pace 1991).

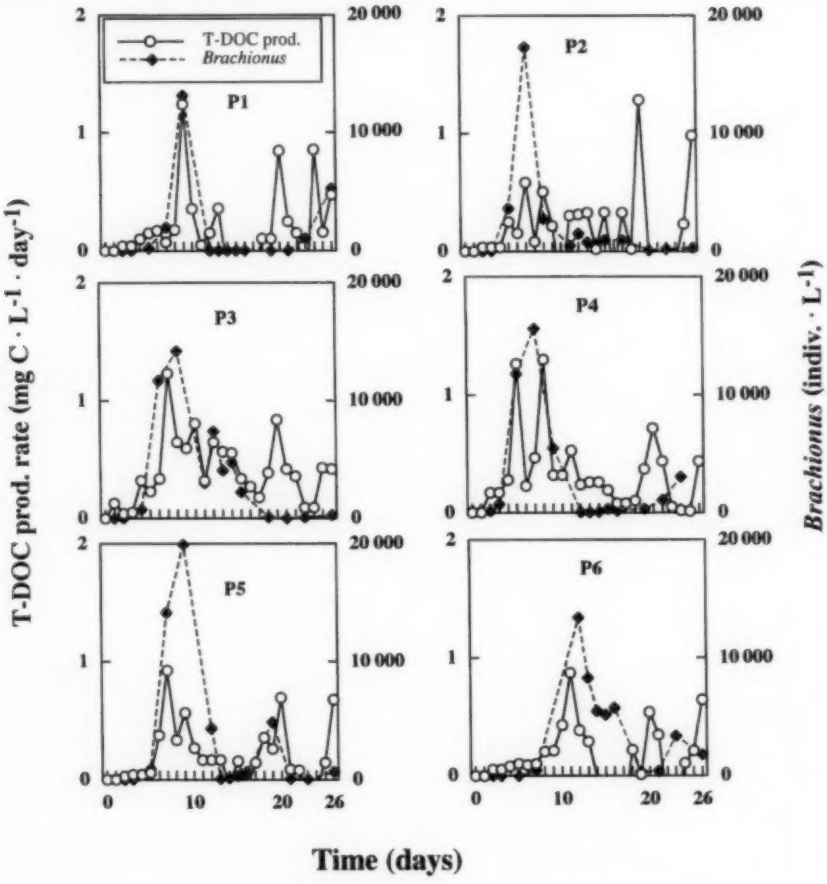
Effect of phytoplankton species composition on DOC concentration changes

Previous studies found that DOC production was related to excretion and lysis during the algal bloom peak and decline (Jensen et al. 1985; Hama and Handa 1987; Munster and Chrost 1990). In the present experiments, we observed the higher T-DOC concentrations during peaks of zooplankton density or biomass. In Lake Kasumigaura, there is no report on the significant DOC production by lysis of cyanobacteria, although the occurrence of microbial agents that cause lysis of cyanobacteria was reported (Yamamoto 1981).

Thus, we tried to differentiate the daily net changes in T-DOC in pond waters caused by zooplankton grazing activity under two different dominant phytoplankton by means of two main factors: daily net change in chlorophyll *a* concentration and daily net change in T-DOC during two different dominant phytoplankton blooms (Fig. 5), suggesting that changes in cyanobacterial chlorophyll *a* concentration were closely

© 1997 NRC Canada

Fig. 7. Daily T-DOC production rates and *B. calyciflorus* densities in ponds P1–P6.



related to those in T-DOC concentration . That is, the decrease in cyanobacterial chlorophyll *a* concentration was related to the increase in daily DOC concentration during the middle stage of these experiments whereas the increase in chlorophyll *a* concentration, active growth of cyanobacteria, did not result in the increase in daily T-DOC concentration during the first stage. In the case of green algae, points were irregularly scattered around the origin and daily change in T-DOC did not show any relationship to daily change in chlorophyll *a* concentration. Although the daily change in T-DOC includes the effects of bacterial mineralization, excretion by phytoplankton, and dilution resulting from inflow water, the relationship between net increase in DOC and decrease in chlorophyll *a* suggests that high T-DOC production was mainly related to changes in cyanobacterial biomass, rather than to those in green algal biomass (Fig. 5).

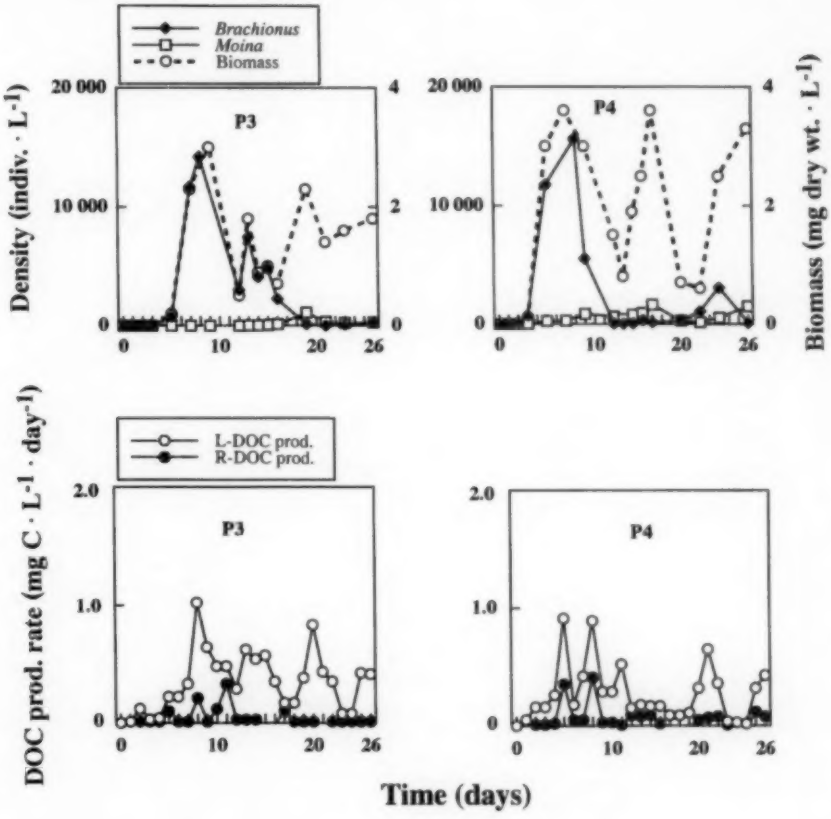
We attributed the rapid decrease in cyanobacterial biomass to zooplankton grazing activity in the middle stage of the experiments. However, there are two other possibilities. The first is that the bloom-forming cyanobacteria were heavily grazed by zooplankton. Fulton and Paerl (1987) reported that *Microcystis* sp. was available as food for large-bodied cladocerans, and Hanazato and Yasuno (1987) confirmed that *Brachionus calyciflorus* could utilize senescent *Microcystis* sp. in Lake Kasumigaura. The second is that the rapid decrease in chlorophyll *a* concentration might have been due to nutrient limitation. In the present experiments, nutrient depletion was never observed during the experimental period (Fig. 6). Thus, it is reasonable to conclude that excretion by *B. calyciflorus* during cyanobacterial blooming may be an important source of T-DOC.

Earlier studies have demonstrated excretion of dissolved organic matter (DOM) by zooplankton (Webb and Johannes 1967; Eppley et al. 1973; Goma et al. 1996). Further, the loss of cellular contents by phytoplankton cell breakage during macrozooplankton grazing has also been observed (Lampert 1978; Copping and Lorenzen 1980). However, these studies used only green algae and diatoms as food for zooplankton.

In the present study, T-DOC concentration rapidly increased during the period of decreasing cyanobacterial biomass, as indicated by phycocyanin concentration (Fig. 2), and scarcely increased during the period of rapid cyanobacterial growth (Fig. 3). However, while green algae were dominant in the late stage of the experiments, no clear relationship between

© 1997 NRC Canada

Fig. 8. Daily L-DOC and R-DOC production rates and zooplankton density and biomass in P3 and P4.



changes in T-DOC and chlorophyll *a* concentrations was discerned.

L-DOC and R-DOC production mechanisms

During the initial 4 days of the experiment, T-DOC production rates were low, less than 0.2 mg $C \cdot L^{-1} \cdot day^{-1}$. The rapid increases in T-DOC production rates in all ponds occurred when the first large chlorophyll *a* peaks declined to near minima (Fig. 2) and the first peak of *B. calyciflorus* density was observed (Fig. 7). The T-DOC production rates ranged from 0.58 to 1.24 mg $C \cdot L^{-1} \cdot day^{-1}$ and *B. calyciflorus* accounted for about 99% of total zooplankton densities in all ponds. After the first peak of *B. calyciflorus* density, at least two peaks in T-DOC production rates also appeared, although no large peak of zooplankton density was observed.

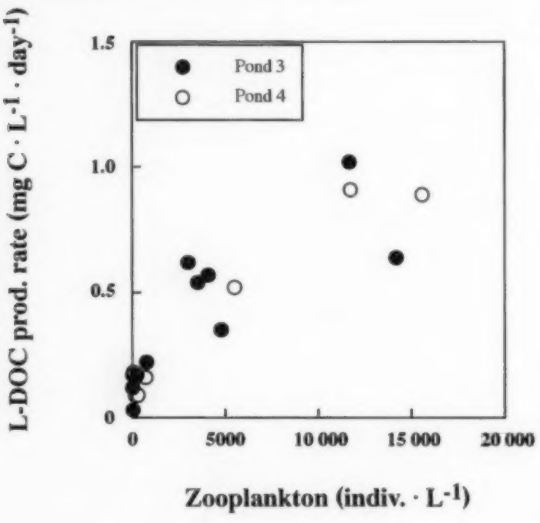
The rapid increases in L-DOC and R-DOC production rates in P3 and P4 also occurred when the first peak of *B. calyciflorus* density was observed (Fig. 8) and chlorophyll *a* concentrations declined to near minima (Fig. 2). L-DOC and R-DOC production rates ranged from 0.04 to 1.02 mg $C \cdot L^{-1} \cdot day^{-1}$ and from 0.01 to 0.41 mg $C \cdot L^{-1} \cdot day^{-1}$, respectively. Although some peaks of T-DOC production rate appeared after the first peak of *B . calyciflorus* density, other zooplankton density peaks were not observed, except for small density peaks by *Moina micrura*. However, small density peaks by *M. micrura* (second dominant) yielded to high biomass peaks because of large size (Fig. 8). Those biomass peaks roughly coincided with relatively large peaks in L-DOC production rate. Thus, it may be possible to explain those peaks by assuming that *M. micrura* excreted L-DOC.

The present study showed that L-DOC concentration fluctuated with changes in zooplankton density (Fig. 8) and revealed a highly significant linear relationship between L-DOC production rates and zooplankton densities ($r = 0.74$, $P < 0.01$) (Fig. 9). The present study also showed that such autochthonously produced L-DOC can be a substrate for bacteria, and L-DOC production via zooplankton excretion may be a major pathway in summer DOC dynamics in eutrophic lakes (Eppley et al. 1973, 1981; Sondergaard et al. 1995).

These results suggested that production of L-DOC and R-DOC was closely related to zooplankton excretion and that most R-DOC production was probably due to the grazing activity of *B. calyciflorus* during the middle stage of the experiment. In previous studies, R-DOC was considered to be composed of humic substances of high molecular weights that were resistant to bacterial attack and its formation required a

© 1997 NRC Canada

Fig. 9. Linear relationship between L-DOC production and zooplankton density in P3 and P4 ($r = 0.74$, $P < 0.01$).



long period (Geller 1985, 1986; Munster and Chrost 1990; Tranvik 1992). The present study revealed, in contrast, that the R-DOC was also produced through zooplankton grazing activity. However, we could not determine whether the R-DOC was directly excreted by zooplankton or not.

The present study suggested that zooplankton excretion can be an important source of T-DOC in eutrophic lakes during summer and that R-DOC can be autochthonously produced during zooplankton grazing activity. Therefore, excretion of T-DOC by zooplankton grazing activity should be added as one of the significant sources of DOC in eutrophic lakes.

Acknowledgments

J-C. Park thanks the Japanese Ministry of Education, Science and Culture for a Scholarship during this work. The authors would like to acknowledge the helpful comments and correction of English on this manuscript by Drs. Y. Aruga and M. Omori, Tokyo University of Fisheries, and R. Weisburd, National Institute for Environmental Studies. Dr. Hanazato, National Institute for Environmental Studies, kindly identified our zooplankton samples.

References

Baines, S.B., and Pace, M.L. 1991. The production of dissolved organic matter by phytoplankton and its importance to bacteria: patterns across marine and freshwater systems. Limnol. Oceanogr. **36**: 1078–1090.

Bratbak, G. 1987. Carbon flow in an experimental microbial ecosystem. Mar. Ecol. Prog. Ser. **36**: 267–276.

Brock, T.D., and Clyne, J. 1984. Significance of algal excretory products for growth of epilimnetic bacteria. Appl. Environ. Microbiol. **47**: 731–734.

Chrost, R.J. 1986. Algal–bacterial metabolic coupling in the carbon and phosphorus cycle in Lakes. *In* Proceedings of the 4th International Symposium of Microbial Ecology. pp. 360–366.

Chrost, R.H., and Faust, M.A. 1983. Organic carbon release by phytoplankton: its composition and utilization by bacterioplankton. J. Plankton Res. **5**: 477–493.

Copping, A.E., and Lorenzen, C.J. 1980. Carbon budget of a marine phytoplankton–herbivore system with carbon-14 as a tracer. Limnol. Oceanogr. **25**: 873–882.

Eppley, R.W., Renger, E.H., Venrick, E.L., and Mullin, M.M. 1973. A study of plankton dynamics and nutrient cycling in the central gyre of the North Pacific Ocean. Limnol. Oceanogr. **18**: 543–551.

Eppley, R.W., Horrigan, S.G., Fuhrman, J.A., Brooks, E.R., Price, C.C., and Sellner, K. 1981. Origins of dissolved organic matter in southern California coastal waters: experiments on the role of zooplankton. Mar. Ecol. Prog. Ser. **6**: 149–159.

Fukushima, T., Park, J.C., Imai, A., and Matsushige, K. 1996. Dissolved organic carbon in a eutrophic lake; dynamics, biodegradability and origin. Aquat. Sci. **58**: 139–157.

Fulton, R.S. III., and Paerl, H.W. 1987. Toxic and inhibitory effects of the blue-green algae *Microcystis aeruginosa* on herbivorous zooplankton. J. Plankton Res. **9**: 837–855.

Geller, A. 1985. Degradation and formation of refractory DOM by bacteria during simultaneous growth on labile substrates and persistent lake water constituents. Schweiz. Z. Hydrol. **47**: 27–44.

Geller, A. 1986. Comparison of mechanisms enhancing biodegradability of refractory lake water constituents. Limnol. Oceanogr. **31**: 755–764.

Goma, R.H., Aizaki, M., Hukushima, T., and Otsuki, A. 1996. Significance of zooplankton grazing activity as a source of dissolved organic nitrogen, urea and dissolved free amino acids in a eutrophic shallow lake: experiments using outdoor continuous flow pond systems. Jpn. J. Limnol. **57**: 1–13.

Hama, T., and Handa, N. 1987. Pattern of organic matter production by natural phytoplankton population in a eutrophic lake. 2. Extracellular products. Arch. Hydrobiol. **109**: 227–243.

Hanazato, T., And Yasuno, M. 1987. Evaluation of *Microcystis* as food for zooplankton in a etrophic lake. Hydrobiologia, **144**: 251–259.

Hobbie, J.E. 1992. Microbial control of dissolved organic carbon in lakes: research for the future. Hydrobiologia, **229**: 169–180.

Jensen, L.M., Jorgensen, O.N.G., and Sondergaard, M. 1985. Specific activity. Significance in estimating release rates of extracellular dissolved organic carbon (EOC) by algae. Verh. Int. Ver. Limnol. **22**: 2893–2897.

Lampert, W. 1978. Release of dissolved organic carbon by grazing zooplankton. Limnol. Oceanogr. **23**: 831–834.

Marker, A.F., Nush, E.A., Rai, H., and Riemann, B. 1980. The measurement of photosynthetic pigments in freshwater and standardization of methods: conclusion and recommendation. Arch. Hydrobiol. Beih. Ergebn. Limnol. **14**: 91–106.

Meili, M. 1992. Sources, concentrations and characteristics of organic matter in softwater lakes and streams of the Swedish forest region. Hydrobiologia, **229**: 23–41.

Middelboe, M., Nielsen, B., and Sondergaard, M. 1992. Bacterial utilization of dissolved organic carbon (DOC) in coastal waters: determination of growth yield. Arch. Hydrobiol. Beih. Ergebn. Limnol. **37**: 51–61.

Munster, V.U. 1985. Investigations about structure, distribution and dynamics of different organic substrates in the DOM of lake Plubsee. Arch. Hydrobiol. (Suppl.), **70**: 429–480.

Munster, U., and Chrost, R.J. 1990. Aquatic microbial ecology. Biochemical and molecular approaches. Springer-Verlag New York Inc., New York. pp. 8–46.

© 1997 NRC Canada

Nixdorf, B. 1990. The fate of phytoplankton primary production: losses in relation to bacterial metabolism in a eutrophic shallow lake. Arch. Hydrobiol. Beih. Ergebn. Limnol. **34**: 61–65.

Ogura, N. 1972. Rate and extent of decomposition of dissolved organic matter in surface seawater. Mar. Biol. **13**: 89–93.

Okada, M., and Aiba, S. 1983. Simulation of water-bloom in a eutrophic lake — II. Reassessment of buoyancy, gas vacuole and turgor pressure of *Microcystis aeruginosa*. Water Res. **17**: 877–882.

Otsuki A., Omi, T., Hashimoto, S., Aizaki, M., and Takamura, N. 1994. HPLC fluorometric determination of natural phytoplankton phycocyanin and its usefulness as cyanobacterial biomass in highly eutrophic shallow lake. Water Air Soil Pollut. **76**: 383–396.

Satoh, Y., and Abe, H. 1987. Dissolved organic matter in colored water from mountain bog pools in Japan. II. Biological decomposability. Arch. Hydrobiol. **111**: 25–35.

Schindler, D.W., Bayley, S.E., Curtis, P.J., Parker, B.R., Stainton, M.P., and Kelly, C.A. 1992. Natural and man-caused factors affecting the abundance and cycling of dissolved organic substances in precambrian shield lakes. Hydrobiologia, **229**: 1–21.

Servais, P., Billen, G., and Hascoet, M.C. 1987. Determination of the biodegradable fraction of dissolved organic matter in waters. Water Res. **21**: 445–450.

Servais, P., Anzil, A., and Ventresque, C. 1989. Simple method for determination of biodegradable dissolved organic carbon in water. Appl. Environ. Microbiol. **55**: 2732–2734.

Sondergaard, M. 1984. Dissolved organic carbon in Danish lakes: concentration, composition, and lability. Verh. Int. Ver. Limnol. **22**: 780–784.

Sondergaard, M., and Borch, N.H. 1992. Decomposition of dissolved organic carbon (DOC) in lakes. Arch. Hydrobiol. Beih. Ergebn. Limnol. **37**: 9–20.

Sondergaard, M., Riemann, B., Jensen, L.M., Jorgensen, N.O.G., Bjornsen, P.K., Olesen, M., Larsen, J.B., Greetz-Hensen, O., Hansen, J., Christoffersen, K., Jespersen, A., Andersen, F., and Bosselmann, S. 1988. Pelagic food web processes in an oligotrophic lake. Hydrobiologia, **164**: 271–286.

Sondergaard, M., Hansen, B., and Markager, S. 1995. Dynamics of dissolved organic carbon lability in a eutrophic lake. Limnol. Oceanogr. **40**: 46–54.

Tranvik, L.J. 1992. Rapid microbial production and degradation of humic-like substances in lake water. Arch. Hydrobiol. Ergebn. Limnol. **37**: 43–50.

Watanabe, Y. 1984. Transformation and decomposition of photosynthetic products of lake phytoplankton. Jpn. J. Limnol. **45**: 116–125.

Webb, K.L., and Johannes, R.E. 1967. Studies of the release of dissolved free amino acids by marine zooplankton. Limnol. Oceanogr. **12**: 376–382.

Wetzel, R.G. 1983. Limnology. Saunders College Publishers, Philadelphia, Pa. pp. 665–706.

Wetzel, R.G. 1992. Gradient-dominated ecosystems: sources and regulatory functions of dissolved organic matter in freshwater ecosystems. Hydrobiologia, **229**: 181–198.

Yamamoto, T. 1981. Observation on the occurrence of microbial agents which cause lysis of blue-green algae in Lake Kasumigaura. Jpn. J. Limnol. **42**: 20–27.

© 1997 NRC Canada

Early maturity in amago salmon (*Oncorhynchus masu ishikawai*): an association with energy storage

Jeffrey T. Silverstein, Hisako Shimma, and Hiroshi Ogata

Abstract: Two groups of amago salmon (*Oncorhynchus masu ishikawai*) with large differences in incidence of early maturity were reared in similar environments and periodically sampled beginning 1 week after exogenous feeding commenced in January and ending in June when maturing individuals could be distinguished. Mean weight of fish in the two groups did not differ beyond the first month of feeding; however, the early-maturing group had significantly greater variation in weight, significantly higher triacylglycerol content in the first sample taken, and maintained higher triacylglycerol content throughout the experiment. Thus, we describe fat storage differences in early-maturing salmon and in salmon that do not mature early, at a far younger age than has been reported previously. In addition, glycogen content and the activities of several enzymes involved in energy storage were examined. Seasonal trends in energy storage show an overall reduction in fat storage in both groups and some differences in glycogen storage dynamics between the groups in late spring. Our results support a previous hypothesis that a size or energy storage threshold must be surpassed for maturation to occur and furthermore suggest that the decision to mature could be made at a very early stage of development.

Résumé : Deux groupes de saumons amago (*Oncorhynchus masu ishikawai*) qui présentent de grandes différences quant à l'apparition de la maturité précoce ont été élevés dans des environnements semblables et échantillonnés sur une période débutant 1 semaine après le commencement de l'alimentation exogène en janvier et se terminant en juin, au moment où les individus commençant à prendre de la maturité pouvaient être distingués. Le poids moyen des poissons appartenant aux deux groupes ne différaient pas après le premier mois suivant le début de l'alimentation exogène; cependant, le groupe présentant une maturité précoce présentait, de manière statistiquement significative, une variation de poids plus grande et une teneur en triacylglycérols plus élevée dans le premier échantillon prélevé et a conservé une teneur en triacylglycérols plus élevée pendant toute la durée de l'expérience. Ainsi, nous avons décrit les différences touchant le stockage des lipides chez les saumons présentant une maturité précoce et les saumons ne présentant pas une telle maturité à un âge beaucoup plus jeune que ce que l'on avait rapporté antérieurement. De plus, la teneur en glycogène et les activités de plusieurs enzymes intervenant dans le stockage des lipides ont été examinées. Les tendances saisonnières dans le stockage des lipides montrent une réduction globale du stockage des lipides dans les deux groupes et une certaine différence dans la dynamique de stockage du glycogène entre les groupes à la fin du printemps. Nos résultats viennent appuyer une hypothèse formulée antérieurement selon laquelle un seuil de taille et de stockage d'énergie doit être franchi pour qu'il y ait maturation et, de plus, laissent entendre que la décision de prendre de la maturité pourrait être prise à un stade de développement très précoce.
[Traduit par la Rédaction]

Introduction

In many salmonids such as brook trout (*Salvelinus fontinalis*) (Alm 1959), coho salmon (*Oncorhynchus kisutch*) (Silverstein and Hershberger 1992), amago salmon (*Oncorhynchus masu ishikawai*) (Shimma et al. 1994), and Atlantic salmon (*Salmo salar*) (Thorpe et al. 1983) the relationship between growth rate and age at maturation is negative, so that faster growing individuals, particularly males, mature at an earlier age. The connection between the somatotropic and gonadotropic axes is undoubtedly strong (LeBail 1988), although its mechanism is not well understood.

The inverse relationship between growth and age at maturation is of great concern to commercial salmon growers. Because salmon virtually stop growing when they begin to mature sexually (Asknes et al. 1986), recently achieved substantial improvements in fish growth performance cannot be realized owing to early maturity (O'Flynn et al. 1992).

Several researchers have documented increased condition factors (weight per unit length) and fat levels in fast-growing, early-maturing males in salmon populations (Bailey et al. 1980; Saunders et al. 1982; Rowe and Thorpe 1990*b*; Silverstein and Hershberger 1992). These observations have led to a hypothesis that some growth or energy storage threshold must be surpassed for sexual maturation to ensue (Garrison 1971; Bailey et al. 1980; Saunders et al. 1982; Herbinger and Newkirk 1990; Rowe et al. 1991; Berglund 1992; Herbinger and Friars 1992; Hankin et al. 1993). The genetic correlation

Received January 16, 1996. Accepted July 15, 1996.
J13253

J.T. Silverstein,[1] H. Shimma, and H. Ogata. National Research Institute of Aquaculture, Nansei, Mie, Japan, 516-01.

[1] Author to whom all correspondence should be sent at the following address: University of Washington, School of Fisheries, 355100, Seattle, WA 98195, U.S.A.
e-mail: jsilver@u.washington.edu

© 1997 NRC Canada

between growth and early maturation has not been estimated, but there is a significant positive phenotypic correlation between growth at a young age and early maturation (Thorpe et al. 1983; Tofteberg and Hansen 1987; Skilbrei 1989; Silverstein and Hershberger 1992). Recent work on Atlantic salmon has shown that fat storage dynamics are a good indicator of maturational schedule, adding support to the size – energy storage threshold hypothesis (Rowe et al. 1991) and encouraging studies on physiological differences between fish that mature early and those that do not.

Unfortunately, work on the physiology and mechanism of early maturity in salmon has been hampered by the circumstance that maturation is a process that begins months before the external signs that indicate maturity are present. It is currently impossible to distinguish individuals as maturing or nonmaturing, for comparative analyses of growth and physiological parameters, during the initial 4–5 months after hatch. Work to date on early maturity in Atlantic salmon was mainly done on fish that matured at 24 months of age without experiencing a seaward migration (cf. Rowe and Thorpe 1990*a*, 1990*b*). Studies aiming to clarify physiological properties differentiating maturing from nonmaturing fish have mainly segregated 12-month-old fish by size, because larger fish are more likely to mature early, or marked and followed a small number of individuals from 12 months of age (Thorpe et al. 1983; Rowe and Thorpe 1990*a*, 1990*b*; Berglund 1992; Simpson 1992).

Our purpose in this study was to determine if and when body fat content diverged between fish that did and did not mature early and to compare the activities of enzymes involved in energy storage and mobilization. We wanted to examine metabolic differences as soon as exogenous feeding began. In our study, we worked on fish in which early maturity occurs at 12 months of age to reduce variation caused by events in previous years. To meet our goal, we established two genetic lines of fish with a large difference in the incidence of early maturity within the same environment and compared the dynamics of energy stores in these lines.

Amago salmon were selected as the experimental animals because lines of amago with different incidences of early maturity that originate from the same parental stock have been identified (Shimma and Kitamura 1987; Shimma et al. 1994; H. Shimma, unpublished data). Early maturity in these fish occurs at 1 year (12 months) of age; the rest of the population matures at 2 years of age.

This method of using distinct genetic lines to examine a trait has been successfully used to understand physiological differences leading to different phenotypes in invertebrate, avian, and mammalian systems (Bulfield 1972; Bishop and Hill 1985; Bulfield et al. 1988; Clark and Keith 1988; Hastings and Hill 1990).

Materials and methods

Experimental animals

Amago salmon gametes were collected from three lines to produce the crosses used in this experiment. First, eggs and sperm were collected at the Gifu Prefectural Fisheries Experimental Station from 2-year-old fish of a line that had been selectively bred for a low incidence of early maturity. Six full-sib families were produced from these gametes, and these families were considered the putative group of fish that did not mature early. Early-maturing (12-month-old) males used to produce the putative early-maturity group were from the line maintained at the National Research Institute of Aquaculture (NRIA; Japan). This line was originally acquired from the Gifu Prefectural Fisheries Experimental Station and subsequently selectively bred for a high incidence of early maturity by one of us (Shimma and Kitamura 1987; Shimma et al. 1994). Females for the early-maturing line were taken from a different strain because the early-maturing females of the NRIA strain have very small eggs; because egg size has been shown to have an effect on age at maturity in some populations (Bilton 1970; Silverstein and Hershberger 1992), this factor was considered important to control. These females were taken from the Misugi strain (Shimma and Kitamura 1987). This strain also has a high incidence of early maturation among males (>80%, J.T. Silverstein and H. Shimma, unpublished data), and because the females were 2 years old the egg size was the same as that in the lines that did not mature early. Five full-sib families were produced by crossing the eggs of Misugi females with the sperm of NRIA males. These families made up the putative high incidence of the early-maturity group.

All crosses were made at the NRIA freshwater hatchery in Tamaki, Japan, between October 29 and November 5, 1993. Each full-sib family was incubated separately and transferred into separate containers for initial rearing. The number of fish moved into rearing containers was the same for all families, and the commercial trout food provided was given at the same ration levels (Leitritz and Lewis 1980) to all groups. When the fish were approximately 6 g, at 5 months post-hatch, all individuals were cold branded (Mighell 1969) to identify family membership and transferred to a single large concrete pond for rearing to July.

Sampling

Beginning at 1 week after feeding was initiated, approximately 1 month post-hatch, samples were taken every 2 weeks for 3 months (January 13, 1994, to March 24, 1994), and thereafter, samples were taken monthly for 3 months (April 7, 1994, to June 30, 1994) until sampling was terminated. At each sampling period, 40 individuals from each family were randomly chosen, anesthetized with MS-222 (Sandoz), and weighed, fork length was measured, and pools of 10 fish (seven fish in monthly samples) were processed for biochemical analyses. Four pools of fish from each family were killed at every sampling period. The fish taken for biochemical analyses were wrapped in plastic or foil and quickly immersed in liquid nitrogen and then placed on dry ice until transfer to a −80°C freezer.

In July when sampling was terminated the incidence of maturity was determined for each family by dissecting at least 50 males in each family and macroscopically examining testicular development. Initially, gonad weights were measured and gonadosomatic index (GSI) was calculated on a subsample of 200 fish across all families. A clear bimodal pattern developed. Fish judged immature had a modal GSI of 0.02–0.03 (range 0.00–0.08) whereas fish judged mature had their major peak at a GSI of 0.20 (range 0.09–0.31). The thickening testes of maturing males were easily distinguished, and thereafter, maturing and nonmaturing males were distinguished on the basis of testicular thickening only. Approximately 140 fish in each family were maintained until September and checked for maturity to confirm our earlier estimates. Including both July and September samples, a minimum of 101 males were examined from each family.

Biochemical analysis

Triacylglycerol (TG) levels, glycogen (GLY) levels, and the enzyme activities of malic enzyme (ME), glucose-6-phosphate dehydrogenase (G6PDH), fatty acid synthetase (FAS), glycogen synthase (GS), and glycogen phosphorylase (GP) were measured in tissue homogenates of pooled samples of amago from each of the high- and low-incidence families over time. In the samples taken between January 13, 1994, and March 24, 1994, whole-body homogenates of the

© 1997 NRC Canada

pooled fish were made by partially thawing the fish and then grinding with a Potter-Elvington homogenizer. A weighed sample of the tissue slurry was then diluted 1:10 (w/v) with homogenization buffer (stopping buffer modified from Moon et al. (1989): 50 mM Tris, 100 mM NaF, 5 mM EGTA, 5 mM EDTA, 5 mM dithiothreitol, and 0.1 mM phenylmethylsulfonylflouride (PMSF)) and thoroughly homogenized in a Teflon homogenizer. All grinding and homogenization steps were carried out on ice. The resulting homogenates were centrifuged (20 min 10 000 × *g* at 4°C) and the supernatants were distributed into 96-well microplates, 25 μL/well. For measurement of TG levels the homogenates were further diluted 1:3 with phosphate-buffered saline before distribution into microplates. Microplates were then frozen at −80°C until analysis.

Samples taken from April 7, 1994, through June 30, 1994, were processed similarly, except that livers were removed from partially thawed fish and only livers were used to make the homogenates.

For all biochemical measurements, the 96-well microplates were removed from the −80°C freezer and allowed to warm to room temperature (23°C) for 30 min.

TG and ME were measured with the procedures of Clark and Keith (1989). For the ME assay, the blanks received incubation medium without malic acid.

GLY was measured by modifying the procedures of Clark and Keith (1989) and Bergmeyer (1963). Briefly, the plates were first floated in a boiling water bath for 1 min and quickly moved to an ice water bath to chill. Then, 75 μL of amyloglucosidase (EC 3.2.1.3) dissolved in acetate buffer (0.2 M; pH 4.8; 75 units/mL) or 75 μL of acetate buffer without amyloglucosidase (for endogenous glucose measurement) was added. After incubation at 37°C for 90 min, absorbance of the plate at 450 nm was read. Subsequently, 100 μL of the Sigma preparation of glucose oxidase and peroxidase (PGO: catalog No. 510-8) with 0.16 mg *o*-dianisidine dihydrochloride/mL was added and plates were incubated for a further 90 min at room temperature. Finally, absorbance at 450 nm was read again and initial values were subtracted from final values. The wells without amyloglucosidase were used to estimate endogenous glucose, and this value was subtracted from the wells with amyloglucosidase to calculate GLY concentration. GLY standards were included on each plate.

G6PDH was measured by the method of Bergmeyer (1963), modified to a microplate technique by reducing sample volumes (to 25 μL) and incubation buffer volumes (to 200 μL). Blanks for this procedure lacked $NADP^+$ in the incubation medium.

FAS was measured by adding 120 μL of incubation medium (20 mM Tris-HCl, 33 μM acetyl-CoA, 100 μM malonyl-CoA, and 100 μM NADPH; pH 7.4) to 25 μL of sample. After a 30-min incubation, absorbance at 340 nm was read five times at 5-min intervals. Blanks received no malonyl-CoA.

GS was measured with the procedure of Sheridan et al. (1985), modified to a microplate technique by reducing sample volumes and media volumes. Twenty-five microlitres of the initial incubation medium (50 mM Tris, 1 mM EDTA, 4 mM cysteine, 10 mg GLY/mL, 3 mM UDP glucose, and 10 mM glucose-6-phosphate; pH 7.8) was added to each well and the plates were incubated for 45 min at room temperature. The reaction was stopped by floating the plate on a boiling water bath for 1 min, followed by quick transfer to an ice water bath. Then, 25 μL of phosphoenolpyruvate (10 mM; in 400 mM KCl and 100 mM $MgCl_2$) and three units of pyruvate kinase were added to each well, followed by a 30-min incubation at 37°C. Next, 25 μL of dinitrophenylhydrazine (0.15% in 2 N HCl) was added, followed by incubation at 37°C for 5 min. Finally, 200 μL of a 1:4 solution of 10 M NaOH − 95% EtOH was added for 40 min. Absorbance at 490 nm was read. Blanks (initial incubation medium added after boiling) and UDP standards were run with each plate.

GP was assayed according to Moon et al. (1989). Two incubation buffers were used, buffers A and B. Both buffers contained 50 mM KH_2PO_4, 15 mM $MgSO_4$, 0.5 mM $NADP^+$, 10 μM glucose-1,6-bisphosphate, 1 mg GLY/mL, 0.8 unit phosphoglucomutase/mL, 6 units G6PDH/mL, and 2 mM AMP. In addition to these components, buffer A also contained 10 mM EDTA and 10 mM caffeine, pH 7.0, to measure activity of the active form of the enzyme, and buffer B contained 0.25 mM EDTA, pH 7.0, to measure total GP activity.

All chemicals used were obtained from either Sigma Chemicals or Wako Chemicals and were the top grades available.

Statistical analyses

Two sample *t* tests were used to compare between groups with high and low incidence of early maturity. The significance level was set at $P \leq 0.05$. Additionally, nested analysis of variance was used to distinguish between groups and families within groups.

Results

Rates of maturation

Of the six putative families that did not mature early, only two had very low rates of maturity. In one family, 9% of the males were mature and in the other, none of the males was mature. Other families ranged between 18 and 34% mature. Therefore, the two families with low rates of maturity were considered the group that did not mature early. In the putative early-maturing families, three of the families had a high incidence of male maturity, 59–67%, and the remaining families were below 36% and not considered in subsequent analyses. Early-maturing females were not observed in any of the families.

Growth

The mean size of the groups that did and did not mature early (Fig. 1A; Table 1) differed significantly in the first two samples, January 13 and 27, in which the group that did not mature early was longer and heavier (Table 1). Thereafter, mean weight did not differ over the course of the experiment. However, the variance in weight was significantly greater ($P < 0.05$) for the early-maturing groups on March 11 and from April 7 to the end of the experiment (Fig. 1B).

Liver weights, measured in the samples from April 7 through June 30, were not different between the stocks until the final sample when the early-maturing group had significantly heavier livers ($P < 0.05$) and significantly higher hepatosomatic indices (HSI) (see Table 1). Post hoc comparison of liver weight and HSI between females, non-maturing males, and maturing males showed that this difference was due to greater liver weights of early-maturing males.

Biochemical measurements

Seasonal fluctuations in TG contents in whole-body and liver samples were similar in the two groups of fish. TG contents in whole-body samples measured from January 13 to March 24 (Fig. 2) were, with the exception of the January 27 sample, significantly greater in the early-maturing group. Also, liver TG contents, measured between April 7 and June 30 (Fig. 3), were significantly greater in the early-maturing group. Activities of enzymes related to fatty acid generation, FAS, ME, and G6PDH, did not consistently differ between the groups (Table 2).

GLY contents (Table 3) in the first month of the study were low, and owing to the small amount of tissue available, values for the groups that did and did not mature early were not compared. Means for the January 27 and February 10 sample, derived by pooling six and nine measurements, respectively,

© 1997 NRC Canada

Table 1. Length, weight, liver weight, and HSI (mean ± SEM) in the group not maturing early (NEM) and the early-maturing (EM) group.

	Length		Weight		Liver weight		HSI	
Date	NEM	EM	NEM	EM	NEM	EM	NEM	EM
Jan. 13	nm	nm	0.154±0.005	0.137±0.002*	nm	nm	nm	nm
Jan. 27	3.43±0.03	3.05±0.03*	0.37±0.01	0.25±0.01*	nm	nm	nm	nm
Feb. 10	4.08±0.04	4.00±0.04	0.68±0.02	0.60±0.02	nm	nm	nm	nm
Feb. 24	5.06±0.08	5.02±0.05	1.38±0.06	1.30±0.04	nm	nm	nm	nm
Mar. 11	5.95±0.07	5.93±0.10	2.26±0.08	2.13±0.10	nm	nm	nm	nm
Mar. 24	6.97±0.10	7.29±0.09*	3.65±0.16	3.99±0.15	nm	nm	nm	nm
Apr. 7	8.27±0.12	8.33±0.14	6.31±0.28	6.47±0.33	0.097±0.004	0.104±0.006	1.69±0.04	1.55±0.04
May 6	11.26±0.16	10.83±0.18	15.43±0.64	13.67±0.68	0.221±0.009	0.203±0.008	1.44±0.03	1.49±0.02
June 2	12.69±0.20	12.97±0.19	21.94±0.99	23.35±1.17	0.331±0.013	0.341±0.015	1.48±0.02	1.44±0.02
June 30	14.68±0.17	14.64±0.23	34.01±1.22	35.79±1.88	0.429±0.018	0.510±0.025*	1.33±0.02	1.41±0.02*

Note: NEM group, n = 60; EM group, n = 90. nm, measurement not made.
*Significant difference between the NEM and EM groups at the 0.05 level.

Fig. 1. Growth of amago salmon over time. (A) Mean weight in the groups maturing early and not maturing early at each sample; error bars represent standard error. (B) Coefficient of variation (standard deviation of weight/mean × 100) for weight at each sample for the two groups.

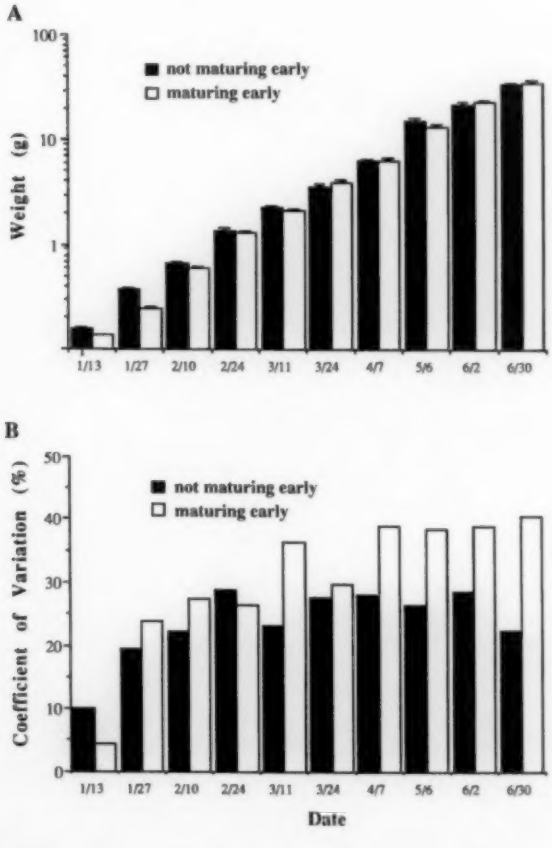


Fig. 2. TG concentration (mean ± SEM) in whole-body homogenates of fish from the groups maturing early and not maturing early (sample sizes in parentheses, each sample is a pool of 10 fish).

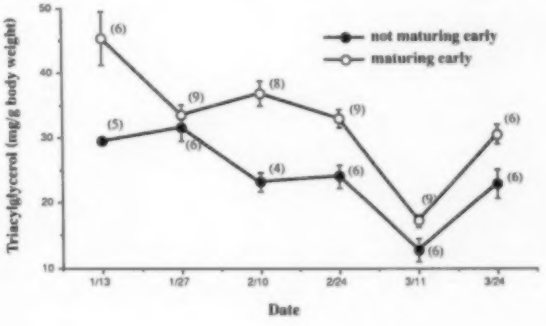
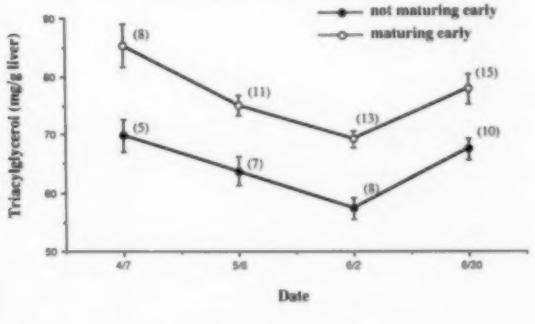


Fig. 3. TG contents (mean ± SEM) in liver homogenates of fish from the groups maturing early and not maturing early (sample sizes in parentheses, each sample is a pool of seven fish).



© 1997 NRC Canada

Table 2. Activities (mean ± SEM) of enzymes involved in lipogenesis in the group not maturing early (NEM) and the early maturing (EM) group.

	FAS (μmol NADPH·g tissue^{-1}·min^{-1})		ME (μmol NADPH·g tissue^{-1}·min^{-1})		G6PDH (μmol NADPH·g tissue^{-1}·min^{-1})	
Date	NEM	EM	NEM	EM	NEM	EM
Jan. 13	nd	nd	0.051±.003	0.043±.002*	0.063±.007	0.060±.005
Jan. 27	nd	nd	0.071±.003	0.056±.002*	0.034±.007	0.036±.003
Feb. 10	nd	nd	0.076±.004	0.072±.003	0.003±.003	0.018±.003*
Feb. 24	nd	nd	0.093±.005	0.096±.004	0.016±.002	0.021±.006
Mar. 11	nd	nd	0.091±.003	0.091±.005	0.009±.003	0.013±.003
Mar. 24	nd	nd	0.099±.002	0.108±.003*	0.042±.007	0.064±.008*
Apr. 7	0.026±.001	0.028±.002	0.267±.027	0.288±.022	0.698±.070	1.047±.041*
May 6	0.023±.002	0.024±.001	0.562±.033	0.582±.023	1.482±.088	1.429±.035
June 2	0.024±.002	0.027±.001	0.600±.038	0.639±.028	1.525±.121	1.591±.067
June 30	0.023±.003	0.022±.002	0.684±.019	0.574±.023*	2.065±.050	1.816±.065*

Note: NEM group, n = 8 pools; EM group, n = 12 pools. nd, not detected. Values are for whole-body tissue from January 27 to March 4 and for liver tissue only from April 7 to June 30.

*Significant difference between NEM and EM groups at 0.05 level.

are shown in Table 2. In the February 24 and March 11 samples, the nonmaturing group had a tendency toward higher GLY values, but the difference was only significant for the March 11 sample. Liver GLY contents determined in the April 7 through June 30 samples showed a very different pattern between groups. The maturing group maintained a steady GLY content throughout this period whereas the nonmaturing group showed a large fluctuation, dropping steeply in May and again at the end of June (Table 3). GLY synthesis and breakdown were assessed by measuring GS activity and GP activity (Table 3). Although some seasonal variation was evident, differences between maturing and nonmaturing groups were not significant. The May and late June drops in liver GLY seen in the nonmaturing group were not closely reflected by significant differences in GS or GP activity, but the higher GLY contents seen in the June 30 sample of the early-maturing group are paralleled by lower total GP activity.

Discussion

We were successful in rearing groups with different incidences of early maturity under similar environmental conditions. The range in the incidence of early maturity was not unexpected considering the low heritability estimates for this trait in other salmonids (Atlantic salmon, Gjerde 1984; coho salmon, Silverstein and Hershberger 1992). Replication of families within groups that did not mature early allowed comparison of size and a suite of biochemical parameters between these groups, beginning at initiation of exogenous feeding many months earlier than in any previous study to date.

The most apparent growth difference between these groups was not average size, but the variance of size. Review of the frequency distributions of fish weight within these groups showed that while the nonmaturing fish had a normal distribution with a single mode for each sample, the early-maturing group had a bimodal distribution, evident by the March 24 sample (data not shown). It was because of this bimodal distribution that the variance in size for the early-maturing group was large. Although not presenting direct evidence, these data suggest that fish maturing at 1 year of age were in the upper mode of the size distribution in the early-maturing group. Our data support the idea that maturing fish are larger in the spring preceding maturity and thus sustain the hypothesis of a size – energy storage threshold that fish must surpass to sexually mature (Saunders et al. 1982; Herbinger and Newkirk 1990; Rowe and Thorpe 1990*a*, 1990*b*; Rowe et al. 1991; Herbinger and Friars 1992; Simpson 1992; Hankin et al. 1993).

Fat content as an indicator of energy storage has been considered an important threshold character limiting early maturity of salmon. The striking difference found in TG contents between the two groups of fish in this study, obvious from the first sample taken 1 week after feeding had begun, provides experimental support for the hypothesis that energy stores have an important role in the decision to mature. Rowe et al. (1991) and Simpson (1992) reported that 24-month-old early-maturing fish were fatter than nonmaturing cohorts months before sexual maturity, and in this study, fish destined to mature at 12 months were fatter much earlier. The difference in fat content within 1 week of exogenous feeding indicates different physiological priorities for energy partitioning in these groups from a very preliminary stage, possibly during incubation. Energy contents of the eggs from both groups were not measured at the beginning of this study, but egg size did not differ between these groups. If fat content is an important characteristic influencing the decision to mature, we cannot rule out the possibility that the fish destined to mature at 1 year of age had been committed to the maturation pathway by the time of our first sample. Additionally, the higher fat content found in the early-maturing group throughout the study suggests important differences in the size of the energy store defended by these groups. Clearly, fat content and size are two parameters associated with early maturity in salmon, but as stated by Simpson (1992), clarification of the respective roles of size and energy storage necessitates independent manipulation of these variables. Studies designed to manipulate these variables independently are underway.

© 1997 NRC Canada

Table 3. GLY contents and activities of enzymes (mean ± SEM) involved in GLY synthesis and breakdown in the group that did not mature early (NEM) and the early-maturing (EM) group.

	GLY (mg·g tissue^{-1})		GS (μmol UDP·min^{-1}·g tissue^{-1})		GP total activity (μmol NADPH·min^{-1}·g tissue^{-1})		GP (% active)	
Date	NEM	EM	NEM	EM	NEM	EM	NEM	EM
Jan. 13			0.173±0.007	0.132±0.010*	0.330±0.008	0.233±0.012*	3.31±0.61	5.27±0.77
Jan. 27	0.62±0.09[†]		0.075±0.010	0.109-±0.014	0.193±0.020	0.200±0.004	7.11±1.12	6.85±0.39
Feb. 10	0.24±0.05		0.047±0.007	0.041±0.003	0.026±0.009	0.059±0.011	71.10±24.51	31.48±7.07
Feb. 24	0.89±0.12	0.52±0.07	0.043±0.003	0.048±0.007	0.060±0.006	0.111±0.028	31.35±5.67	26.58±7.32
Mar. 11	2.14±0.46	0.85±0.11*	0.064±0.007	0.052±0.007	0.033±0.005	0.083±0.013*	58.17±12.00	39.36±8.41
Mar.24	2.34±0.39	3.77±1.19	0.065±0.022	0.106±0.026	0.144±0.047	0.254±0.058	17.81±5.85	9.49±1.42
Apr. 7	30.42±1.48	25.71±0.83	0.288±0.012	0.329±0.036	0.084±0.009	0.167±0.019*	67.12±9.51	70.90±4.13
May 6	11.03±1.55	23.07±1.83*	0.482±0.035	0.459±0.035	0.348±0.038	0.309±0.020	73.59±3.56	74.81±2.23
June 2	25.24±1.15	23.74±1.53	0.598±0.029	0.549±0.018	0.315±0.014	0.307±0.018	64.27±2.93	61.99±1.70
June 30	14.30±2.04	23.40±1.84*	0.572±0.033	0.501±0.039	0.170±0.018	0.129±0.013*	42.93±5.09	38.25±9.17

Note: NEM group, n = 8 pools; EM group, n = 12 pools. Values are for whole-body tissue from January 27 to March 4 and for liver tissue only from April 7 to June 30. GP total activity refers to activity in buffer B (see Materials and methods). The GP ratio expresses the percentage of GP present in the active form calculated as GP active (buffer A) / GP total × 100.

*Significant difference between the NEM and EM groups at 0.05 level.

[†]GLY values for January 27 and February 10 were not calculated separately for each group.

GLY contents have never been compared between salmon that mature early and those that do not. The lower level of GLY storage in the whole body of early-maturing fish on March 11 may indicate a preference for fat storage in this group. The most noticeable difference in GLY contents between the group maturing early and the group not maturing early over the course of this experiment was the spring fluctuation seen in liver GLY of the nonmaturing group versus the stability of the maturing group. Differences in life history could be responsible for the May drop in liver GLY of the nonmaturing group. Smoltification was not measured; however, the nonmaturing group would be expected to have more smolting individuals than the early-maturing group (Thorpe 1986, 1987), and smolting salmon typically show reduced liver glycogen (Woo et al. 1978; Plisetskaya et al. 1988; Mayer et al. 1994). Amago salmon are known to smoltify in the autumn; however, there appears to be a small rise in circulating thyroxine levels and hypoosmoregulatory ability in the spring (Fujioka et al. 1990), suggesting a pseudo or vestigal smolting event in the spring. Such an event may also explain the seasonal drop in liver TG content seen in this study and shown to be typical of smolting salmonids (Sheridan et al. 1983; Sheridan 1985).

The differences seen in the fat contents between salmon maturing early and those not maturing early (Rowe et al. 1991; Simpson 1992) led us to investigate lipogenic activity to determine if the difference in fat storage was accompanied by differences in lipogenesis. Although some measures in isolated samples were significantly different, no consistent pattern of difference emerged. Several of the enzymes monitored in this study were evaluated in coho salmon by Lin et al. (1997*b*) as indicators of hepatic lipogenesis in response to fasting. The liver appeared to be by far the most important site of lipogenesis (Lin et al. 1997*a*; review by Greene and Selivonchick 1987); nevertheless, a substantial metabolic perturbation (23-day fast) was required to detect a change in lipogenic enzyme activity. Therefore, the lack of difference in activity in this experiment is perhaps not surprising. For a better understanding of the fat dynamics, comparison of lipolytic activity should be included in future studies. Nevertheless, the positive association between lipid contents and maturation strategy was clear, although the mechanism by which the different lipid store sizes are maintained remains to be clarified. Difference in metabolic rate, or food consumption within groups, could account for observed differences in fat storage.

There were both strengths and weaknesses in this approach to physiological comparison of amago salmon maturing early and those not maturing early. Our strategy allowed early detection of the differences in body fat, thus supporting hypotheses of a size – energy storage threshold postulated for much older salmon, and further, suggested that characteristics defined by first feeding may influence maturation schedule. Additionally, a variety of other parameters not yet monitored could be investigated using this paradigm. The major weakness of this method was the necessity of pooling many individuals for sampling, and thus losing individual information.

The families identified in this study as having high and low incidences of early maturity were marked and their offspring will be available for subsequent studies of early maturity. By using these offspring, we hope to achieve greater uniformity in the maturation phenotypes within these two groups, giving more power to future investigations.

Acknowledgements

We are grateful to the Gifu Prefecture Fisheries Experimental Station for supplying us with gametes for the fish not maturing early that were used in this study. Prof. Erika Plisetskaya

© 1997 NRC Canada

greatly improved earlier versions of this paper. J.T.S. was supported during this project by the STA Fellowship of the Japanese Science and Technology Agency and the United States National Science Foundation. Additional support during the writing of this article was provided by U.S. Department of Agriculture grant No. 94-37206-1096.

References

Alm, G. 1959. Connection between maturity, size and age in fishes. Rep. Inst. Freshwater Res. Drottningholm, **40**: 5–145.

Asknes, A., Gjerde, B., and Roald, S.O. 1986. Biological, chemical and organoleptic changes during maturation of farmed Atlantic salmon, *Salmo salar*. Aquaculture, **53**: 7–20.

Bailey, J.K., Saunders, R.L., and Buzeta, M.I. 1980. Influence of parental smolt age and sea age on growth and smolting of hatchery-reared Atlantic salmon (*Salmo salar*). Can. J. Fish. Aquat. Sci. **37**: 1379–1386.

Berglund, I. 1992. Growth and early sexual maturation in Baltic salmon (*Salmo salar*) parr. Can. J. Zool. **70**: 205–211.

Bergmeyer, H.U. 1963. Glycogen determination with amyloglucosidase. *In* Methods of enzymatic analysis. Academic Press, New York. pp. 1127–1131.

Bilton, H.T. 1970. Maternal influence on the age of maturity of Skeena river sockeye salmon (*Oncorhynchus nerka*). Tech. Rep. Fish. Res. Board Can. No. 167.

Bishop, S.C., and Hill, W.G. 1985. Effects of selection on growth, body composition and food intake in mice. III. Correlated responses: growth body composition, food intake and efficiency and catabolism. Genet. Res. **46**: 57–74.

Bulfield, G. 1972. Genetic control of metabolism: enzyme studies of *obese* and *adipose* mutants in the mouse. Genet. Res. **20**: 51–64.

Bulfield, G., Isaacson, J.H., and Middleton, R.J. 1988. Biochemical correlates of selection for weight-for-age in chickens: twenty-fold higher muscle ornithine decarboxylase levels in modern broilers. Theor. Appl. Genet. **75**: 432–437.

Clark, A.G., and Kieth, L.E. 1988. Variation among extracted lines of *Drosophila melanogaster* in triacylglycerol and carbohydrate storage. Genetics, **119**: 595–607.

Clark, A.G., and Kieth, L.E. 1989. Rapid enzyme kinetc assays of individual *Drosophila* and comparisons of field-caught *D. melanogaster* and *D. simulans*. Biochem. Genet. **27**: 263–277.

Fujioka, Y., Fushiki, S., Tagawa, M., Ogasawara, T., and Hirano, T. 1990. Seasonal changes in plasma thyroxine levels in Biwa and amago salmon reared in the pond. Nippon Suisan Gakkaishi, **56**: 249–254.

Garrison, R.L. 1971. Effect of rapid smolt growth on coho maturation. J. Wildl. Manage. **35**: 762–766.

Gjerde, B. 1984. Response to individual selection for age at sexual maturity in Atlantic salmon. Aquaculture, **38**: 229–240.

Greene, D.H.S., and Selivonchick, D.P. 1987. Lipid metabolism in fish. Prog. Lipid Res. **26**: 53–85.

Hankin, D.G., Nicholas, J.W., and Downey, T.W. 1993. Evidence for inheritance of age of maturity in chinook salmon (*Oncorhynchus tshawytscha*). Can. J. Fish. Aquat. Sci. **50**: 347–358.

Hastings, I.M., and Hill, W.G. 1990. Analysis of lines of mice selected for fat content. 2. Correlated responses in the activities of enzymes involved in lipogenesis. Genet. Res. **55**: 55–61.

Herbinger, C.M., and Friars, G.W. 1992. Effects of winter temperature and feeding regime on the rate of early maturation in Atlantic salmon (*Salmo salar*) male parr. Aquaculture, **101**: 147–162.

Herbinger, C.M., and Newkirk, G.F. 1990. Sources of family variability for maturation incidence in cultivated Atlantic salmon. Aquaculture, **85**: 153–162.

LeBail, P.Y. 1988. Growth reproduction interaction in salmonids. *In* Reproduction in fish: basic and applied aspects in endocrinology and genetics. Institut national de recherches agronomiques, Paris, France. pp. 91–106.

Leitritz, E., and Lewis, R.C. 1980. Trout and salmon culture. Calif. Dep. Fish Game Fish Bull. No. 164.

Lin, H., Romsos, D.L., Tack, P.I., and Leveille, G.A. 1977*a*. Influence of dietary lipid on lipogenic enzyme activities in coho salmon (*Oncorhynchus kisutch* (Walbaum)). J. Nutr. **107**: 846–854.

Lin, H., Romsos, D.L., Tack, P.I., and Leveille, G.A. 1977*b*. Effects of fasting and feeding various diets on hepatic lipogenic enzyme activities in coho salmon (*Oncorhynchus kisutch* (Walbaum)). J. Nutr. **107**: 1477–1483.

Mayer, I., Borg, B., and Plisetskaya, E.M. 1994. Plasma levels of insulin and liver glycogen contents in one- and two-year old Atlantic salmon (*Salmo salar* L.) during the period of parr–smolt transformation. Fish Physiol. Biochem. **13**: 191–197.

Mighell, J.L. 1969. Rapid cold-branding of salmon and trout with liquid nitrogen. J. Fish. Res. Board Can. **26**: 2765–2769.

Moon, T.W., Foster, G.D., and Plisetskaya, E.M. 1989. Changes in peptide hormones and liver enzymes in the rainbow trout deprived of food for 6 weeks. Can. J. Zool. **67**: 2189–2193.

O'Flynn, F.M., Friars, G.W., and Bailey, J.K. 1992. Development of a selection index to improve market value of cultured Atlantic salmon (*Salmo salar*). Genome, **35**: 304–310.

Plisetskaya, E.M., Swanson, P., Bernard, M., and Dickhoff, W.W. 1988. Insulin in coho salmon (*Oncorhynchus kisutch*) during the parr to smolt transformation. Aquaculture, **72**: 151–164.

Rowe, D.K., and Thorpe, J.E. 1990*a*. Suppression of maturation in male Atlantic salmon parr by manipulation of feeding and growth in spring months. Aquaculture, **86**: 291–313.

Rowe, D.K., and Thorpe, J.E. 1990*b*. Differences in growth between maturing and non-maturing male Atlantic salmon, *Salmo salar* L., parr. J. Fish Biol. **36**: 643–658.

Rowe, D.K., Thorpe, J.E., and Shanks, A.M. 1991. Role of fat stores in the maturation of male Atlantic salmon (*Salmo salar*) parr. Can. J. Fish. Aquat. Sci. **48**: 405–413.

Saunders, R.L., Henderson, E.B., and Glebe, B.D. 1982. Precocious sexual maturation and smoltification in male Atlantic salmon (*Salmo salar*). Aquaculture, **28**: 211–229.

Sheridan, M.A. 1985. Changes in the lipid composition of juvenile salmonids associated with smoltification and premature transfer to saltwater. Aquaculture, **45**: 387–388.

Sheridan, M.A., Allen, W.V., and Kerstetter, T.H. 1983. Seasonal variations in the lipid composition of steelhead trout, *Salmo gairdneri*, associated with the parr–smolt transformation. J. Fish Biol. **23**: 125–134.

Sheridan, M.A., Woo, N.Y.S., and Bern, H.A. 1985. Changes in the rates of glycogenesis, glycogenolysis, lipogenesis, and lipolysis in selected tissues of the coho salmon (*Oncorhynchus kisutch*) associated with parr–smolt transformation. J. Exp. Zool. **236**: 35–44.

Shimma, H., and Kitamura, H. 1987. Maturation of female yearling of amago salmon *Oncorhynchus rhodurus* and acquisition of fertilized eggs. Yoshoku Kenkyusho Kenkyu Hokoku, **12**: 93–96.

Shimma, H., Kagawa, H., and Hirose, K. 1994. Effect of fingerling size on the precocious maturation in amago salmon. Yoshoku Kenkyusho Kenkyu Hokoku, **23**: 55–63.

Silverstein, J.T., and Hershberger, W.K. 1992. Precocious maturation in coho salmon (*Oncorhynchus kisutch*): estimation of the additive genetic variance. J. Hered. **83**: 282–286.

Simpson, A.L. 1992. Differences in body size and lipid reserves between maturing and non-maturing Atlantic salmon parr, *Salmo salar*, L. Can. J. Zool. **70**: 1737–1742.

Skilbrei, O.T. 1989. Relationships between smolt length and growth and maturation in the sea of individually tagged Atlantic salmon (*Salmo salar*). Aquaculture, **83**: 95–108.

© 1997 NRC Canada

Thorpe, J.E. 1986. Age at first maturity in Atlantic salmon, *Salmo salar*: freshwater period influences and conflicts with smolting. *In* Salmonid age at maturity. *Edited by* D.J. Meerburg. Can. Spec. Publ. Fish. Aquat. Sci. No. 89. pp. 7–14.

Thorpe, J.E. 1987. Smolting versus residency: developmental conflict in salmonids. Am. Fish. Soc. Symp. **1**: 244–252.

Thorpe, J.E., Morgan, R.I.G., Talbot, C., and Miles, M.S. 1983. Inheritance of developmental rates in Atlantic salmon, *Salmo salar* L. Aquaculture, **33**: 119–128.

Tofteberg, P., and Hansen, T. 1987. Relationship between age at maturity and growth rate in farmed rainbow trout, *Salmo gairdneri*. *In* Proceedings of the World Symposium on Selection, Hybridization, and Genetic Engineering in Aquaculture. *Edited by* K. Tiews. Heenemann Verlagsgesellschaft, Berlin, Germany. pp. 405–420.

Woo, N.Y.S., Bern, H.A., and Nishioka, R.S. 1978. Changes in body composition associated with smoltification and premature transfer to seawater in coho salmon (*Oncorhynchus kisutch*) and king salmon (*Oncorhynchus tschawytscha*). J. Fish Biol. **13**: 421–428.

© 1997 NRC Canada

A test of recruitment limitation and the potential for artificial enhancement of spiny lobster (*Panulirus argus*) populations in Florida

Mark J. Butler IV and William F. Herrnkind

Abstract: We conducted a field experiment in the Florida Keys in which we manipulated Caribbean spiny lobster (*Panulirus argus*) settlement and shelter for juveniles to (*i*) investigate how these factors influence local recruitment and (*ii*) evaluate the efficacy of such practices proposed to enhance spiny lobster populations. The number of small juvenile lobsters significantly increased at six 0.05-ha sites where 12 artificial shelters were added, but was unchanged on three unmanipulated sites. Adding over 150 new settlers to three of the shelter-supplemented sites did not measurably increase juvenile lobster abundance above that attributable to shelter enhancement alone. Mark–recapture results confirmed that the observed increase in small juveniles on shelter-supplemented sites was not due to immigration. The addition of artificial structures to nursery sites appears to have reduced predation on the vulnerable small juveniles and led to their local retention. In contrast, larger, more mobile juveniles were no more abundant where shelter was added than on the unmanipulated sites. These results support the proposition that local recruitment of *P. argus* may be increased by augmenting natural shelter with appropriately designed artificial structures, but the broader implications of enhancing lobster populations on the surrounding natural communities are unknown.

Résumé : Nous avons réalisé une expérience de terrain dans les Cayes de la Floride dans laquelle nous avons manipulé la fixation et les abris des langoustines juvéniles de l'espèce *Panulirus argus* pour (*i*) étudier comment ces facteurs influent sur le recrutement local et (*ii*) évaluer l'efficacité de ces pratiques proposées pour accroître la population de langoustine. Le nombre de langoustines juvéniles de petite taille a augmenté de manière statistiquement significative dans six sites de 0,05 ha où l'on avait ajouté 12 abris artificiels, mais il est demeuré inchangé dans trois sites non manipulés. Le fait d'ajouter 150 nouveaux pionniers dans trois des sites dans lesquels on avait ajouté des abris n'a pas fait augmenter de manière mesurable l'abondance des langoustines juvéniles au-delà de l'augmentation attribuable à la seule augmentation du nombre d'abris. Les résultats des expériences de marquage–recapture ont confirmé que l'augmentation observée du nombre des juvéniles de petite taille dans les sites où l'on avait ajouté des abris n'était pas attribuable à l'immigration. L'addition de structures artificielles dans les sites d'élevage semble avoir réduit la prédation sur les juvéniles de petite taille vulnérables et avoir favorisé leur implantation locale. Par opposition, les juvéniles plus gros et plus mobiles n'étaient pas plus abondants dans les sites où l'on avait ajouté des abris que dans les sites non manipulés. Ces résultats appuient l'hypothèse que le recrutement local de *P. argus* peut être accru si l'on ajoute aux abris naturels des structures artificielles bien conçues, mais les conséquences à plus grande échelle de l'accroissement de la population de langoustines sur les communautés naturelles environnantes sont inconnues.
[Traduit par la Rédaction]

Introduction

Overexploitation and deterioration of the world's coastal fisheries have galvanized fishery scientists and ecologists into an increasingly unified (Richards and Bohnsack 1987; Fritz et al. 1990; Sale 1990; Rosenberg et al. 1993) effort to study recruitment and population dynamics of marine species (Rothschild 1986; Roughgarden et al. 1988; Underwood and Fairweather 1989; Fairweather 1991). Extreme variance in recruitment (i.e., the number of new individuals within a population that survive to a fishable size) characterizes most finfish and invertebrate fisheries (Sissenwine 1984), and understanding the processes involved has emerged as a focus of contemporary marine research. Traditionally, such research has emphasized the theoretical relationship between larval and adult stages, but after years of study, predictions of recruitment based on these relationships have been largely unsatisfying (Sissenwine 1984). Larval transport mechanisms and theoretical adult–larval relationships remain important issues (Victor 1986; Miller et al. 1988; Lewin 1986; Hughes 1990; Possingham and Roughgarden 1990; Fogarty et al. 1991; Rothschild et al. 1996), but settlement and early post-settlement processes that influence recruitment now also demand attention.

For species with complex life cycles (i.e., species with two or more developmental stages requiring spatially separated habitats), understanding recruitment requires information on larval settlement and on ontogenetic shifts in habitat

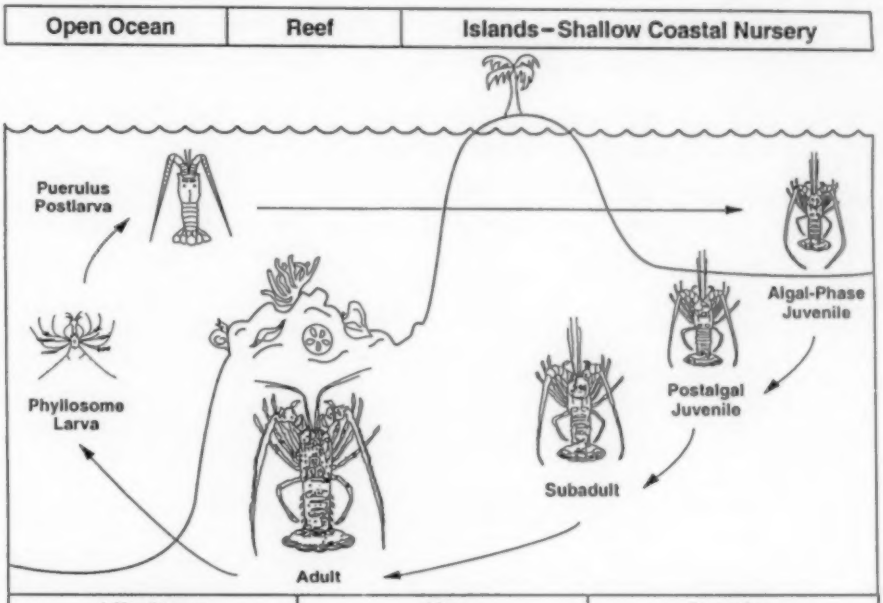
Received January 9, 1996. Accepted July 31, 1996.
J13249

M.J. Butler IV.[1] Department of Biological Sciences, Old Dominion University, Norfolk, VA 23529-0266, U.S.A.
W.F. Herrnkind. Department of Biological Science, Florida State University, Tallahassee, FL 32306-2043, U.S.A.

[1] Author to whom all correspondence should be addressed.
e-mail: mjb100f@viper.mgb.odu.edu

© 1997 NRC Canada

Fig. 1. Diagrammatic summary of the life history of the Caribbean spiny lobster (*Panulirus argus*), including the size, duration, and habitat requirements of each life stage.



Life Stage	Size	Duration
Phyllosome Larva	0.5 - 12 mm CL	9 - 12 Months ?
Puerulus Postlarva	5 - 7 mm CL	2 - 6 Weeks ?
Algal-phase Juvenile	5 - 15 mm CL	2 - 5 Months
Postalgal-phase Juvenile	15 - 45 mm CL	6 - 18 Months
Subadult	45 - 80 mm CL	5 - 9 Months
Adult	80 - >200 mm CL	>10 Years

requirements, shelter availability, and other factors that alter postsettlement survival and growth. Studies are therefore needed that emphasize research on life stages occurring in coastal waters and the linkage between late-stage larval (or postlarval), juvenile, and adult stages. From a practical perspective, resource managers cannot greatly affect oceanic transport of larvae or be certain that they can influence management decisions in other nations whose adult stocks may supply larvae to their own. But managers can act to protect and potentially to enhance nursery habitats essential for postlarval settlement and subsequent juvenile development.

Life history and fishery for the Caribbean spiny lobster

The Caribbean spiny lobster (*Panulirus argus* Latreille) is one of the most heavily fished and commercially significant shellfish throughout its range, from Bermuda to southern Brazil. In Florida, spiny lobsters are the target of intensive commercial trapping (nearly 1 million traps for an average annual catch of 2.5×10^6 kg) and recreational sport-diving fisheries (estimated at 20–25% of commercial harvest; Hunt 1994). The fishery is extensively overcapitalized and is estimated to reduce the adult population by up to 90% annually (Harper 1991; Hunt 1994).

Panulirus argus has a complex life cycle requiring three distinct habitats: coral reef, open ocean, and shallow, vegetated coastal zone (Fig. 1). Mating and spawning occur exclusively along coastal margins near coral or rocky reefs that are accessible to the open ocean. The long-lived (an estimated 9–12 months; Lewis et al. 1952; Kittaka 1994) planktonic larvae may be transported thousands of kilometres, and genetic evidence suggests that there is a single pan-Caribbean stock (Lyons 1980; Silberman and Walsh 1994). The larvae metamorphose offshore into nonfeeding, strongly swimming puerulus postlarvae that are transported inshore during new moon (Little 1977; Butler and Herrnkind 1991; Forcucci et al. 1994) to macroalgae-covered hard-bottom habitat and seagrass where they settle and metamorphose (Marx and Herrnkind 1985; Herrnkind and Butler 1986; Field and Butler 1994). In the Florida Keys, juvenile spiny lobsters exhibit three ecologically distinct phases following settlement: algal phase, postalgal phase, and subadult (Fig. 1).

The algal-phase juveniles (= early benthic phase; sensu Wahle and Steneck 1991; 5–15 mm carapace length (CL)) remain for a few months in vegetation, where they are sheltered from predators (Herrnkind and Butler 1986; Smith and Herrnkind 1992) and have abundant prey (Marx and Herrnkind 1985; Herrnkind et al. 1988). Upon reaching 15–20 mm CL, the juveniles emerge from the macroalgae and, as postalgal phase juveniles (20–45 mm CL), hide during the day under crevices provided by rocks, sponges, octocorals, or

© 1997 NRC Canada

other structures; at night, they forage on small mollusks and crustaceans in the surrounding habitat (Andree 1981; Herrnkind et al. 1994). Large sponges that have deep grooves or are undercut are the most commonly used shelter throughout Florida Bay (73% of the occupied structures; M. Butler and W. Herrnkind, unpublished data). Yet, the proportion of crevice-bearing sponges, hence availability of shelter for juvenile lobsters, varies remarkably among hard-bottom areas.

Small postalgal juveniles roam only a few metres from their daytime shelter (Herrnkind and Butler 1986; Yoshimura and Yamakawa 1988; R. Ramsdell and M. Butler, unpublished data), but at about 45 mm CL (approximately 1 year after settlement), they become nomadic and forage widely in hard-bottom and seagrass habitats (Herrnkind 1980, 1983). As they approach maturity about 2 years after settlement (approximately 76 mm CL), they migrate seaward to the reef tract.

Limitations to Caribbean spiny lobster recruitment

The influx of postlarval lobsters into the Florida Keys and elsewhere in the Caribbean has been monitored using floating collectors that mimic a vegetated substrate (Little 1977; Little and Milano 1980; Heatwole et al. 1992; Forcucci et al. 1994; Herrnkind and Butler 1994). Using similar devices, Australian fishery researchers have been able to correlate the numbers of postlarvae on inshore collectors with commercial catches several years later, and they have used this relationship successfully to predict harvest of western rock lobster (*Panulirus cygnus*) (Phillips 1986; Caputi and Brown 1993). No attempt has been made to equate postlarval influx with harvest in Florida, partly because of inadequate time-series information on postlarval influx and adult harvest. However, the processes of lobster recruitment in Florida and Western Australia are also quite dissimilar. In contrast with the situation in Western Australia, the influx of postlarvae in Florida occurs each lunar cycle (with high variance) and the primary settlement habitat is heterogeneous and ephemeral, which may obviate straightforward stock prediction (Marx 1986; Butler et al. 1995, 1997). Multiple regression analyses linking postlarval influx, benthic settlement, and postalgal juvenile abundance have yielded weak results, but of all of the factors tested, the availability of settlement and postsettlement habitat has explained the greatest variation in local recruitment (Field and Butler 1994; Herrnkind and Butler 1994). This suggests that the number of lobsters that recruit into the postalgal juvenile stage in an area is not a simple reflection of the number of postlarvae settling there. Early postsettlement mortality may decouple the relationship between postlarval supply and juvenile or adult stocks, creating a recruitment or demographic bottleneck, as has been argued for the American lobster (*Homarus americanus*) (Caddy 1986; Wahle and Steneck 1991) and other crustaceans (Steger 1987; Eggleston and Armstrong 1995).

Increasing effort has been directed toward artificial enhancement of lobster fisheries, accomplished through habitat alterations and, to a lesser extent, by supplementary recruitment through stocking (National Fisherman 1986; Addison and Bannister 1994). These efforts follow on the heels of long-standing practices of shelter enhancement in artisanal fisheries but are increasing rapidly, often with governmental support (Stone et al. 1991). In the United States, interest in fisheries enhancement led to the passage of the National Fishing Enhancement Act in 1984 and development of the National Artificial Reef Plan in 1985. The practical intent of such programs is efficient harvest as well as stock enhancement. Although the effectiveness of such measures in increasing short-term harvest is unchallenged, their usefulness as a technique for enhancing populations has rarely undergone rigorous testing, and the premise of enhancement has been strongly challenged (Bohnsack and Sutherland 1985; Bohnsack 1989, 1991; Polovina 1991). Knowledge of the ecological dynamics regulating natural recruitment for most target species is simply absent, so many enhancement efforts become little more than uncontrolled, trial and error manipulations.

In several Caribbean lobster fisheries, artificial structures (e.g., casitas) have been installed in an effort to bolster lobster populations (Cruz et al. 1986; Tangley 1987), but they attract mainly subadult and adult lobsters (Scarratt 1968; Cruz et al. 1986; Eggleston et al. 1990), which raises the possibility that they render the species more vulnerable to fishing, a criticism of artificial reefs generally (Bohnsack 1989). Although such structures reduce local predation on subadults (Eggleston et al. 1990), there is no evidence that recruitment is increased through the nurturing of new settlement or reduction of early postsettlement mortality.

In this study, we tested whether nursery habitat can support substantially larger numbers of juvenile spiny lobsters. We augmented both the numbers of newly settled lobsters and shelter for postalgal juveniles at several field sites and compared the resultant numbers of juveniles at manipulated sites with those at unmanipulated sites. We hypothesized that if recruitment of spiny lobsters is strongly limited by the availability of shelter for later juvenile stages, then the survival or retention of juveniles should increase where shelter is augmented. However, if recruitment were dictated primarily by postlarval supply, then juvenile lobster abundances should increase even more where settlement is artificially enhanced along with shelter.

Methods

Site description and selection

Our experiment was conducted near Old Dan Bank, a shoal in south-central Florida Bay, 2 km north of Long Key, Florida. In May 1988, we selected nine hard-bottom sites for our experiment, each approximately 0.05 ha in area and 1.5–2 m deep. The sites were scattered over approximately 3 km^2 of sea floor. Each site was surrounded by seagrass meadows (*Thalassia testudinum*) and covered by patches of red macroalgae (primarily *Laurencia* spp.) interspersed with sponges and octocorals. Postalgal juvenile lobsters prefer macroalgae-covered hard bottom over seagrass, do not commonly traverse large expanses of sand or seagrass (Herrnkind and Butler 1986), and remain within a few metres of where they settle (Yoshimura and Yamakawa 1988; Norman et al. 1994; Yoshimura et al. 1994; R. Ramsdell and M.J. Butler, unpublished data). Thus, we were reasonably certain that the configuration of our sites would insure their statistical independence. Nonetheless, throughout the study, we monitored the abundance of tagged lobsters found on each site to estimate the degree of intersite movement by lobsters and verify the assumption of site independence.

We documented the initial conditions at our experimental sites by conducting a quantitative survey of the general habitat structure and a pretreatment census of the resident postalgal lobster population structure at each site. The habitat was characterized as the percent cover of any structure (i.e., sand, sponges, octocorals, solution holes, seagrass, and red macroalgae) spanning >10 cm along four haphazardly distributed, nonoverlapping transects within each site. Transects were

© 1997 NRC Canada

6–23 m long, depending on the shape of the hard-bottom site. The pretreatment lobster census was performed using the same technique as in our regular censuses, which are described below.

Experimental design and sampling protocol

After the pretreatment lobster census, we randomly assigned each of the nine sites to one of three treatment conditions (three replicates per treatment): (*i*) natural, unmanipulated control (control), (*ii*) addition of shelter suitable for postalgal juvenile lobsters (shelter addition), and (*iii*) addition of shelter plus the addition of newly metamorphosed algal-stage juvenile lobsters (shelter + settlement addition). We did not independently evaluate the effect of added settlement alone. An additional treatment where settlement alone was manipulated would have been desirable in that it would have yielded a balanced 2 × 2 factorial design, but the need for adequate replication along with limited availability of personnel and the logistical constraints posed by this replicated field manipulation precluded it.

Shelter was increased by haphazardly distributing 12 artificial structures throughout the experimental site. Each artificial structure consisted of two stacked concrete partition blocks; each block was 10.2 × 20.3 × 40.6 cm and contained three 4 × 10 cm holes. The artificial structures incorporated several features characteristic of shelters preferred by spiny lobsters, namely dark, recessed dens with multiple entrances approximately one body length deep (Davis 1985; Spanier and Zimmer-Faust 1988; Eggleston et al. 1990). We have found (M.J. Butler and W.F. Herrnkind, unpublished data) that the average number of juvenile lobsters that dwell in a single artificial shelter is within 5% of the number that is typically found associated with occupied loggerhead sponges. Furthermore, mortality for lobsters dwelling in our artificial shelters is similar to that for lobsters residing under loggerhead sponges (Childress and Herrnkind 1994). Therefore, we are reasonably sure that the addition of this particular type of artificial shelter to experimental sites is equivalent to increasing the density of suitable shelters, such as sponges, both in terms of the lobster carrying capacity and probability of survival in each type of shelter. On the basis of estimates obtained from four 2 m wide belt transects per site conducted at the experimental sites following the study, we estimate that we increased the overall density of structure on each shelter-supplemented site by 25%.

We added first-stage juveniles to three sites during daylight each month (May–September 1988) by releasing newly metamorphosed algal-phase lobsters (5–7 mm CL) individually into large clumps of *Laurencia* spp. Postlarvae were obtained from Witham-type collectors deployed on the oceanside margins of Long Key Channel, which is the natural source of postlarvae for the study area (Herrnkind and Butler 1994). Postlarvae were held in the laboratory in a flow-through seawater table under ambient light until they metamorphosed (about 7 days) into first-stage benthic juveniles, at which time they were released in the field. The number of juvenile lobsters added to each site ranged from 10 to 40/month (mean = 26/month), depending on the magnitude of natural postlarval influx each month; a total of 159 juveniles were added to each of the three shelter + settlement addition treatment sites during the study. At the time of the study, no reliable methods were available for monitoring natural settlement. However, we subsequently estimated settlement using benthic postlarval collectors and by comparing recapture rates of microwire-tagged settlers with those of untagged, natural settlers collected from sites in the region surrounding Old Dan Bank (Herrnkind and Butler 1994; Butler et al. 1997). Those results indicate that we probably increased ambient settlement levels by about 25%. Therefore, structure density and settlement density were increased by roughly similiar amounts.

We censused all lobsters at the sites approximately biweekly from May through September 1988 (experiment 1). Lobsters were censused during the day by SCUBA divers who searched every potential shelter (e.g., sponges, octocorals, coral heads, solution holes, and artificial structures) within each site. Lobsters <45 mm CL were captured whereas those >45 mm CL were only counted; our capture success rate (i.e., number of lobsters sighted that were caught) exceeded 98%. We also recorded the sex, carapace length (nearest 0.1 mm), and the shelter type from which each lobster was collected. Lobsters 20–45 mm CL were marked with coded antenna tags for assessment of potential offsite movement. Antenna tags are external and do not affect survival, but they are lost during ecdysis and so remain useful only until the lobsters' next molt (approximately 4–6 weeks for individuals 20–45 mm CL; Lellis and Russell 1990; Forcucci et al. 1994). All captured lobsters were returned to the same shelter or shelter type on the site.

Estimates of juvenile *P. argus* growth (Lellis and Russell 1990; Phillips et al. 1992; Forcucci et al. 1994) indicate that it takes at least 6–8 weeks for settlers to reach the minimum size (15 mm CL) at which they emerge from settlement habitat and can be accurately censused while in their crevice dens. Thus, we anticipated at least a 6- to 8-week lag before any treatment effects could be detected.

As a separate test of juvenile lobster population responses to shelter addition without the influence of artifically enhanced settlement, we censused the lobsters at each of the nine sites three times the following year (1989) in June, July, and August (experiment 2). Because the artificial structures were left in place since May 1988, we assumed that any persisting treatment effects on postalgal stage lobsters were attributable to added shelter, not to supplemented settlement. These surveys began at least 9 months after we last added first-stage lobsters and 8 months after the last census period. Thus, sufficient time had elapsed during the sampling hiatus (September 1988 – May 1989) to insure that (*i*) few if any juveniles present during the previous year's sampling would still be present on a particular site and (*ii*) the first-stage juvenile lobsters last introduced into the experimental sites had grown large enough either to vacate the sites or to be easily discriminated from recent settlers on the basis of size (Lellis and Russell 1990). During this second sampling period, we tagged all lobsters (except recently molted ones) that were 30–40 mm CL with sphyrion anchor tags inserted in the dorsolateral extensor muscle between the cephalothorax and abdomen. These tags are retained through several molts, have negligible effects on growth, and have been used successfully to mark juveniles (Forcucci et al. 1994; Moriyasu et al. 1994).

Statistical analyses

Initial conditions and suitability of artifical shelters

Initial differences among groups of sites that would later constitute experimental treatment groups were tested (one-factor multivariate analysis of variance, MANOVA; factor = treatment groups) with respect to six independent variables: abundance of small juvenile lobsters (<35 mm CL), abundance of larger juvenile and subadult lobsters (>35 mm CL), and percent cover (arcsin square-root transformed) of sand, seagrass, red macroalgae, and sponges or octocorals. The sponge and octocoral categories, reflecting typical postalgal lobster shelter, were combined, and the solution-hole category (the least abundant type of shelter) was omitted to permit sufficient degrees of freedom for the MANOVA.

We evaluated whether our artificial shelters were used by lobsters in a manner similar to natural postalgal shelters by comparing the numbers and sizes of lobsters captured in artificial shelters and in natural dens. Lobsters were captured from six different shelter types (artificial shelters, macroalgal clumps, solution holes, octocorals, sponges, and sponge–octocoral complexes). Size-specific differences in shelter use were analyzed with a one-factor ANOVA and a Ryan–Einot–Gabriel–Welsh multiple F test (REGWF), for comparisons of specific treatment means.

Treatment effects

For the first experiment conducted in 1988, we evaluated the relative importance of added shelter and increased settlement in affecting

© 1997 NRC Canada

local recruitment of postalgal juveniles by comparing (two-factor repeated measures ANOVA followed by a REGWF test) the number of small (<35 mm CL) juvenile lobsters in each treatment collected during the eight censuses made in 1988 (May–September). The data were log-transformed so that variances among treatment groups were homogeneous ($P > 0.05$, F-max test) and two of the three treatment groups were normally distributed ($P > 0.05$, Shapiro–Wilk test). To be conservative, we did not consider lobsters >35 mm CL in this key analysis because mobility increases with size, and we could not be certain whether lobsters >35 mm CL had immigrated to the site or resulted from local recruitment.

The design of our 1988 experiment did not permit us to evaluate the effect of increased settlement independent of shelter density because the design lacked a treatment where settlement alone was supplemented. The consequence of this is that we could not statistically test for the presence of an interaction between shelter and settlement addition. That is, if adding shelters to a site somehow canceled the effect of settlement enhancement, then we would be unable to distinguish the respective effects of these two separate manipulations. We could, however, subjectively detect a positive interaction (i.e., synergistic effect) between settlement and shelter because the number of lobsters in the shelter + settlement addition treatment would be greater than that in the shelter addition treatment alone.

When we recensused the sites in 1989, only two treatment conditions were still in effect (i.e., control and shelter addition), since settlement enhancement had long since been terminated. Nevertheless, we analyzed the data in the same manner as for the 1988 data and included all three treatment groups, recognizing the prior experimental histories of the sites. The hypothesis addressed in this analysis was whether juvenile lobster (<35 mm CL) abundance on sites supplied with artificial shelters (i.e., the former shelter addition and the shelter + settlement addition treatments) for a full year was greater than their abundance on unmanipulated control sites. The test of this hypothesis is a single orthogonal F test comparing the mean juvenile lobster abundances at shelter addition and shelter + settlement addition sites versus control sites. These results from 1989 provided no information on settlement effects other than a subjective comparison (during the same months of the year: June–August) with results from the 1988 experiment when settlement had been manipulated.

Mark–recapure

The tagging methods we used in 1988 (antenna tags on juveniles 20–45 mm CL but lost at ecdysis) differed from those used in 1989 (sphyrion tags in juveniles 30–40 mm CL and retained through ecdysis), so these data were analyzed separately and were used to address different questions. First, we evaluated the assumption that our sites were independent by determining whether lobsters tagged on one site move to another. Second, we sought to corroborate earlier findings that indicate that small juvenile spiny lobsters display stronger fidelity to a locale than larger juveniles. We did this by comparing (*i*) the proportion of small (20–34 mm CL) and large (35–45 mm CL) juveniles recaptured each census and (*ii*) the proportion of lobsters (by size-class) that were tagged in each census (recapture) sample. We hypothesized that the immigration and emigration (i.e., movement) of large juveniles exceeds that of small juveniles and also exceeds the loss of small juveniles from a site that is due to natural mortality. If so, fewer large juveniles would be recaptured than small ones.

We also used the 1988 and 1989 mark–recapture data to better substantiate whether the effects of our enhancement of shelter and postlarval settlement on the population density of small juveniles (20–34 mm CL) can be attributed to the greater survival and retention of juveniles. Since the 1989 data are based on tags that are retained through ecdysis, Jolly–Seber mark–recapture analysis was possible. Perhaps of greatest interest is the among-treatment comparison of estimated survival probability. Higher estimates of "survival" (= survival – emigration) on the manipulated sites (shelter + settlement addition and shelter addition) compared with the unmanipulated control sites would be consistent with the hypothesis that our manipulations enhanced survival and retention of local recruits.

The 1988 data would not yield reliable Jolly–Seber estimates owing to high rates of tag loss, but they were used to indirectly assess the local recruitment enhancement hypothesis in the following way. If there is a greater decrease in recaptures on control sites than on manipulated sites, then the missing recaptures on control sites either died or emigrated. That is, there was poorer local recruitment in natural areas than in those where shelter or settlement were enhanced. To conclude that small lobsters were remaining on our manipulated sites and were not immigrating into them to seek shelter, the proportion of small marked juveniles (20–34 mm CL) found each census on manipulated sites should vary less through time than the proportion found marked on control sites.

At each biweekly census, we also searched artificial structures that were widely scattered about the seagrass surrounding the sites. We expected that transient marked and unmarked lobsters would appear at these structures depending on the amount of immigration and emigration by lobsters in the area.

Results

Initial conditions and suitability of artificial shelters

Our experimental sites appear to be typical of those in Florida Bay with respect to nursery habitat structure and juvenile lobster abundance. The mean density of all structures >20 cm in diameter (i.e., potential lobster shelters such as sponges, solution holes, octocorals, and coral heads) on our experimental sites was 18 structures/100 m^2 (SD = 7), which is similar to a regional mean of 22 structures/100 m^2 (SD = 44) that we subsequently observed at nearly 200 hard-bottom sites spread throughout the Florida Keys from Biscayne Bay to Key West (W.F. Herrnkind and M.J. Butler, unpublished data). The initial lobster densities at our sites (mean = 6.3 lobsters/site) were also similar to the regional mean density (8.2 lobsters/site).

The experimental sites, grouped by treatment, were also not significantly different from one another with respect to initial conditions, including the number of small lobsters <35 mm CL per site, the number of large lobsters >35 mm CL per site, and the percent bottom cover of four major shelter types (F value for Wilk's lambda = 12.01; df = 12, 2; $P = 0.08$). The univariate tests for these same parameters confirm that none of the dependent variables differed significantly among sites (range of $P = 0.15$–0.91).

The artificial shelters we used in our experiments proved to be an effective facsimile of natural postalgal lobster shelters. Artificial shelters were frequently inhabited by juvenile lobsters. On average, 76% (SD = 16.4) of the postalgal-stage lobsters (<35 mm CL) found on sites supplied with artificial shelters were in those shelters, although the percentage fluctuated among sites and sampling dates. Lobsters found in the artificial shelters were not significantly different in size (mean CL = 32.1 mm) from those found sheltering under octocorals (mean CL = 38.9 mm), sponges (mean CL = 32.9 mm), and octocoral–sponge complexes (mean CL = 32.1 mm). Larger, subadult lobsters usually occupied solution holes (mean CL = 47.6 mm) whereas algal clumps harbored the smallest individuals (mean CL = 20.8 mm); lobsters from these two shelter types differed significantly in size from those in artificial shelters, octocorals, sponges, and octocoral–sponge complexes ($F = 77.64$; df = 5, 1205; $P < 0.001$).

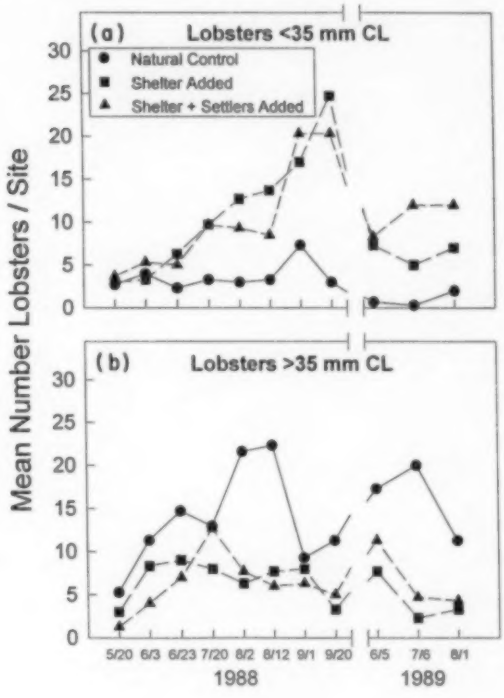
© 1997 NRC Canada

Table 1. Results of two-factor repeated measures ANOVAs analyzing data for the 1988 and 1989 experiments testing the effect of experimental treatment and date on the abundance of small (<35 mm CL) juvenile spiny lobsters at sites in Florida Bay, Fla.

Source	df	MS	*F*	*P*
Experiment 1: 1988				
Treatment	2	9.69	6.92	0.028
Site (treatment)	6	1.39	3.88	0.004
Date	7	1.83	5.11	<0.001
Treatment × date	14	0.34	0.94	0.523
Error	41	0.36		
Experiment 2: 1989				
Treatment	2	8.12	7.18	0.026
Control versus shelter added	1	46.69	389.08	<0.001
Site (treatment)	6	1.13	9.55	<0.001
Date	2	0.16	1.34	0.299
Treatment × date	4	0.12	0.97	0.457
Error	12	0.12		

Note: The 1988 ANOVA was followed by a REGWF test to discern differences among treatment means: control, 3.4; shelter + settlement addition, 11.0; shelter addition, 11.8 (the last two values were not significantly different from each other at $P(\alpha) = 0.05$). A single orthogonal *F* test was conducted for the 1989 data (shown in the ANOVA table) to test whether small juvenile lobster abundance differed significantly between the control treatment versus the two other treatments where shelter for lobsters was increased.

Fig. 2. Mean numbers of postalgal (<35 mm CL; (*a*)) and larger juvenile and subadult (>35 mm CL; (*b*)) spiny lobsters plotted as a function of treatment and census date ($N = 3$ sites/treatment). Census dates during the first experiment in 1988 (May–September) are separated from those of the second experiment conducted in 1989 (June–August) by a break in the axes.



Treatment effects

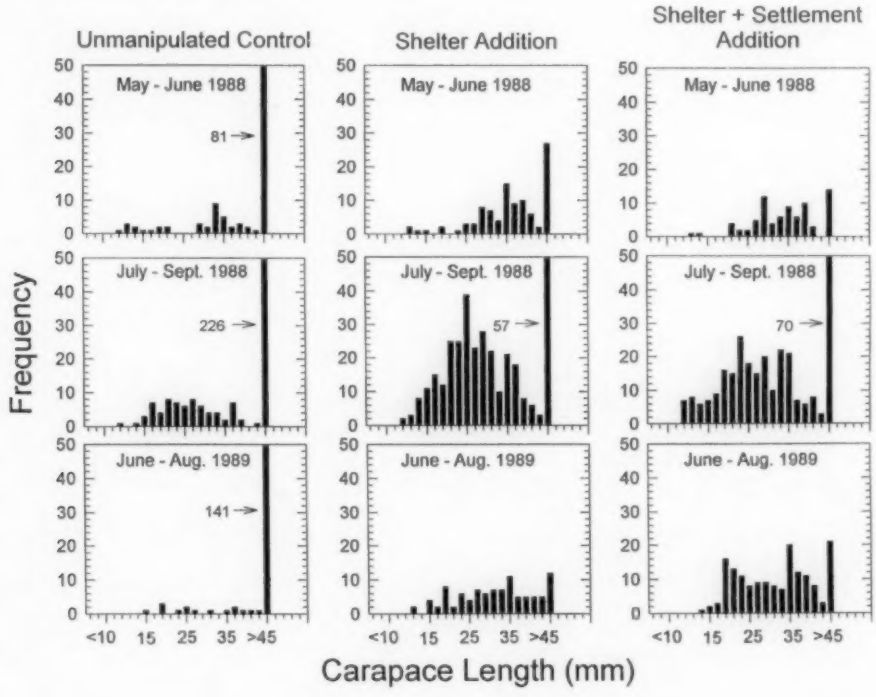
During the first experiment in 1988, small juvenile lobsters (<35 mm CL) became more than three times as abundant in the shelter addition (mean = 11.8 lobsters/site) and shelter + settlement addition treatments (mean = 11.0 lobsters/site) than in the natural control treatment (mean = 3.8 lobsters/site; Fig. 2), which is a significant difference (Table 1). The abundance of small lobsters also differed among sampling dates (Table 1). In 1989, 9 months after we had terminated the settlement enhancement treatment, small juvenile lobsters (<35 mm CL) remained significantly more abundant on sites with artificial shelters (Table 1; Fig. 2). Although the number of small lobsters in all treatments may appear to have been lower in 1989 than in 1988, there was in fact no difference when data from the same season (June–August) were compared (Fig. 2). The late-summer increase in juvenile abundance probably reflects the typically high influx of postlarvae into the Florida Keys during the winter and early spring and their subsequent slow growth at cooler temperatures, which presumably results in a large, broadly overlapping cohort of postalgal juveniles in the summer (Forcucci et al. 1994). In summary, these results indicate that by increasing the number of structures for postalgal lobsters by about 25%, we increased the local abundances of small postalgal lobsters (<35 mm CL) nearly threefold. However, increasing settlement by a similar amount had no effect on mean juvenile abundance beyond the effect that can be attributed to the addition of shelter alone.

The time course of population growth and coincident lobster size distributions suggest that the significant increase in the numbers of juveniles on sites where shelter was added was due to local enhancement of recruitment (i.e., survival of settlers) and not to attraction of small juvenile lobsters (<35 mm CL) from adjacent areas. Size frequency distributions for each treatment over time show that the number of very small individuals on the shelter addition and shelter + settlement addition sites increased only after the expected delay for postsettlement growth and transition from the algal phase (Fig. 3). In contrast, few small lobsters were found on control sites, although the number of large juveniles (>35 mm CL) on those sites fluctuated greatly (Figs. 2 and 3). However, those large immigrant lobsters did not contribute to the significant treatment effect we observed because that analysis was only conducted on small lobsters (<35 mm CL).

The number of large juvenile lobsters (>35 mm CL) observed during the experiment has no direct bearing on the key hypothesis we wished to test concerning local recruitment for two reasons. First, their abundance is also strongly influenced by immigration and emigration, so we cannot use them as reliable indicators of local recruitment. Second, they prefer solution holes and large crevices and rarely used the small

© 1997 NRC Canada

Fig. 3. Size frequency distributions for juvenile spiny lobsters censused under three experimental treatment conditions (natural control, shelter addition, and shelter + settlement addition; $N = 3$ sites/treatment) during three time periods: May–June 1988 (experiment 1: preresponse period), July–September 1988 (experiment 1: response period), and June–August 1989 (experiment 2). Size frequency patterns, instead of the actual magnitudes, are important in these comparisons because the number of censuses differs among time periods. Note the large increase in the abundance of postalgal juveniles in the two treatments where shelter was manipulated during the last two time periods (i.e., response periods for experiments 1 and 2) compared with that in the unmanipulated control treatment.



artificial shelters we deployed. We noted, however, that the mean number of large juveniles (>35 mm CL) was significantly lower on the experimental sites with artificial shelter than on our control sites where the abundance of large juveniles fluctuated dramatically, and this result warrants some explanation. As it turns out, the high mean abundance of large juveniles and subadults on our control sites is due entirely to the presence of two large solution holes (preferred shelter for large juveniles) that existed on two of the three control sites and nowhere else; 79% of the lobsters found on these two sites were in these two solution holes. We also suspect that the temporally variable number of large juveniles on the sites is a consequence of their nomadism and temporary occupancy of these few large solution holes. When the lobsters that sheltered in these two solution holes are omitted from the analysis, the number of large juveniles does not differ among treatments.

Mark–recapture

Intersite movement of antenna-tagged lobsters during the eight biweekly censuses in 1988 was virtually nonexistent. Out of 1171 tagged lobsters and 137 recaptures at the experimental sites, only one individual was not on the site where it was tagged. As expected, small juvenile lobsters (20–34 mm CL) generally remained on the sites longer than large juveniles (35–45 mm CL), as indicated by the higher percentage of small lobsters that were recaptured (mean = 17%) as compared with large lobsters (mean = 8.6%) (Table 2). In addition, searches of widely scattered artificial structures scattered in the seagrass around the sites yielded only 11 lobsters, nearly all of which were >35 mm CL, and only 1 that was tagged and had moved from the nearest site about 10 m away.

The more frequent emigration and immigration of larger, nomadic juveniles on and off the sites led to greater fluctuations in their abundance and, therefore, greater variance in the percentage of large juveniles found tagged each census (mean CV = 158) than small juveniles found tagged (mean CV = 82) (Table 2). The percentage of small juveniles found tagged each census also fluctuated less on sites where shelter was enhanced than on control sites (Table 2). This suggests that movement (emigration and immigration) of small juveniles declined wherever shelter was supplemented.

The mark–recapture results also indicate that survival and retention of small juveniles were higher on sites where shelter was enhanced than on control sites (Table 2). The mean values for recapture success (i.e., percent recaptured per census) and the percentage of the censused population that was found tagged each census were twice as large on manipulated sites as on control sites (Table 2). The lower recapture success for small, less mobile juveniles on control sites suggests that their survival was poorer there. The Jolly–Seber estimates of the

© 1997 NRC Canada

Table 2. Results of the mark–recapture analyses for juvenile lobsters of various size-classes marked with antenna tags in 1988 (experiment 1) and sphyrion tags in 1989 (experiment 2).

Experiment 1: 1988

	Lobsters 20–34 mm CL			Lobsters 35–45 mm CL		
Treatment	% recaptured each census (mean ± SD)	% found tagged each census (mean ± SD)	CV	% recaptured each census (mean ± SD)	% found tagged each census (mean ± SD)	CV
Settlement + shelter	19.8±14.4	24.1±16.3	68	21.8±21.2	18.0±10.6 59	
Shelter	20.9±15.3	23.2±18.1	78	1.5±2.6	2.0±4.5	218
Control	10.3±13.8	12.2±12.4	101	2.4±4.2	4.8±9.5	198
Mean	17.0	19.8	82	8.6	8.3	158

Experiment 2: 1989

	Lobsters 30–40 mm CL	
Treatment	Probability of survival (phi) (mean ± SE)	Population size (*N*) (mean ± SE)
Settlement + shelter	0.78±0.31	27.2±8.7
Shelter	0.67±0.39	18±10.8
Control	0.18±0.05	13.3±2.8

Note: The results are presented separately for each year and for the three experimental treatments, with three replicate sites per treatment. Values estimated for 1988 are based on eight census dates whereas those for 1989 are based on three census dates. The probability of survival and population size estimates in 1989 are based on a Jolly–Seber model analysis.

probability of survival of juveniles 30–40 mm CL in 1989 confirm the 1988 tagging results. The probability of survival (phi) of this lobster size-class on the control sites was only 25% of that on sites where shelter had been enhanced.

Discussion

Roles of shelter and settlement in controlling spiny lobster recruitment

Supplementing shelter for juvenile spiny lobsters at sites in the Florida Keys with suitable settlement habitat significantly increased the local abundance of small postalgal juveniles. We found no evidence that immigration by small juveniles from outside the experimental sites could have caused this effect. Supporting our experimental results, the size structure of the lobster populations and our mark–recapture results reveal that (*i*) small lobsters show strong site fidelity and move little until they exceed 35 mm CL and (*ii*) more algal-phase juveniles survived the transition to the postalgal stage on sites where we supplemented shelter. We suspect that in many areas, natural structures are few or of inappropriate architecture and do not provide effective shelter from the suite of large predators that feed on small (15–25 mm CL) juveniles. Predation is much higher on exposed algal-phase juveniles than on older postalgal juveniles (Smith and Herrnkind 1992), and sponges or other structures do not universally provide protection equal to that afforded by macroalgae (Childress and Herrnkind 1994). Thus, the risk of predation is highest for small juveniles and those that wander in the open to find a protective crevice. By providing more shelter at some sites, we presumably reduced juvenile mortality and the tendency for juveniles to leave those sites. If so, more lobsters would have accumulated on those sites over time. This is not an effect of attracting and accumulating transient, mobile individuals from other areas as can occur with older, more nomadic juveniles (Davis 1985; Eggleston et al. 1990). In our study, large nomadic juveniles were no more abundant where the small artificial shelters were placed than where they were not. If our interpretations are correct, providing refuge tuned to the most vulnerable early benthic life phase, which ordinarily suffers very high predation, potentially increases recruitment to the juvenile population.

On the basis of observations of size-specific habitat use and shelter availability, Wahle and Steneck (1991) provided a similar interpretation of factors limiting *H. americanus* recruitment in New England. They argued that the availability of cobble substrate (and its associated crevices) for settlement and early benthic phase juvenile shelter represents a demographic bottleneck for these populations. In a study of shelter and recruitment of stomatopods, Steger (1987) likewise concluded that the abundance of rock crevices offering protection from predators limited stomatopod populations in coastal Panama. In that system, crevice availability is inversely related to crevice size, and crevice size is directly related to the size of stomatopod that can occupy the space. By following seasonal pulses in stomatopod recruitment through time, Steger (1987) noted the density-dependent damping of settlement peaks as individuals grew larger and encountered increasingly limited shelter availability. The situation for Caribbean spiny lobsters seems similar, but somewhat more complex because of ontogenetic shifts in the types of shelters used (e.g., macroalgae to crevices) and because lobsters attain some degree of size

© 1997 NRC Canada

refuge from predation (Smith and Herrnkind 1992). The results of these studies and others (e.g., Hacker and Steneck 1990; Norman et al. 1994; Eggleston and Armstrong 1995) on different crustaceans in remarkably disparate habitats all point to the importance of adequate structural refuge for early recruits in determining local population density. The implications of these results for adult stocks that are fished, although seemingly obvious, are not easily tested in a manipulative manner. However, a correlation between structural complexity, presumably reflecting the availability of shelter, and adult stock has been shown for at least one species of spiny lobster, the Hawaiian spiny lobster (*Panulirus marginatus*) (Parrish and Polovina 1995).

We did not anticipate that our addition of settlers would be ineffective in boosting the local abundance of juvenile lobsters. The number of postalgal lobsters was identical on sites where either shelter alone or shelter and settlement were supplemented (i.e., mean of 11.8 lobsters/site versus 11.0 lobsters/site, respectively). Also, the results of the 1989 experiment, where only shelter was manipulated, were nearly identical to those obtained in 1988 when both shelter and settlement were increased. Thus, at the levels that we supplemented settlement and shelter, we detected an effect attributable only to increased shelter availability.

This does not imply that fluctuations in settlement magnitude are unimportant in determining the strength of recruitment, and we make this case elsewhere (Herrnkind and Butler 1994; Butler et al. 1997). We suspect that the relative influence of settlement and shelter availability on juvenile recruitment is probably not equivalent and that their effects are not necessarily additive. However, early postsettlement mortality is estimated to exceed 95% (Herrnkind and Butler 1994; Butler et al. 1997), and our supplementation may have been insufficient to be detected. Forcucci et al. (1994) inferred that a tripling in natural settlement was needed to produce a 50% increase in recruitment of postalgal stage juveniles at a shelter-rich site in Florida Bay. Thus, a seemingly moderate increase in sheltering structure for postalgal-stage spiny lobsters may result in large increases in local recruitment whereas a similar recruitment response would require a very large increase in settlement (i.e., larval supply).

The importance of postsettlement processes in changing ontogenetic spatial distributions, although well known for sessile marine invertebrates (Keough and Downes 1982; Fairweather 1988), is not widely appreciated in mobile marine animals (but see Steger 1987). Although it is instructive to contrast the relative importance of larval supply and nursery habitat structure in regulating recruitment, the two probably interact to control Caribbean spiny lobster population structure. The survival of spiny lobsters that settle in nursery areas profuse with shelter appears to be higher than where shelter is scarce. In these areas, settlement magnitude is likely to be directly reflected in recruitment to later juvenile stages several months later. However, mass settlement may not necessarily result in more lobsters in an area if there is insufficient shelter to sustain the later juvenile stages. For example, shelter density at a site near Fiesta Key, <5 km from our Old Dan Bank sites, is among the highest we have recorded in surveys of nearly 200 sites throughout the Florida Keys (M.J. Butler and W.F. Herrnkind, unpublished data). There, the correlation between postlarval supply and recruitment to the postalgal juvenile stage 8 months later was significant ($r = 0.83$; $P < 0.001$; Forcucci et al. 1994). That is, fluctuations in the local recruitment of lobsters in the 30–35 mm CL size-class tracked the fluctuations in settlement that occurred 8 months earlier. However, at our sites near Old Dan Bank, shelter for postalgal stage lobsters is only a third of that available at the nearby Fiesta Key site. Postlarval supply and postalgal juvenile abundance (lagged by 8 months) were not significantly correlated at our Old Dan Bank sites ($r = 0.48$; $P > 0.05$; Butler et al. 1996), even though our study was conducted during the same period of time and probably subject to a postlarval supply similar to that at Fiesta Key. Our experimental data and these examples suggest that local recruitment will only reflect settlement magnitude where shelter for later juvenile stages is sufficiently abundant. Otherwise, the relationship is decoupled, presumably by postsettlement mortality and emigration associated with the lack of shelter for postalgal-stage lobsters.

Another potentially important factor affecting the recruitment of juvenile *P. argus* is the abundance of macroalgal cover available for settling postlarvae (Butler et al. 1997). Unlike the shelters used by postalgal juveniles, the availability of macroalgal settlement habitat is ephemeral both seasonally and regionally in Florida Bay (Butler et al. 1997; Zieman et al. 1989). Thus, the shifting availability of macroalgal nursery habitat coupled with patchy postlarval settlement is likely to result in a locally unpredictable pattern of settlement (Herrnkind and Butler 1994). We could not reliably control the abundance of macroalgae or natural settlement in the experiments described here, although we have shown that they were similar among sites and consistent with levels common in the region.

Artificial enhancement measures

Evaluation of the potential for artificial enhancement of Caribbean spiny lobster populations has centered on two distinct juvenile phases, the algal to postalgal period (this study) and the vagile subadult phase (Cruz et al. 1986; Eggleston et al. 1990). Individuals in the latter phase are the primary occupants of casitas or pesqueros, large artificial structures a few metres across placed in shallow nurseries by lobster fishermen in Central America, Cuba, and the Bahamas (Briones et al. 1994; Cruz and Phillips 1994). Set in seagrass beds where natural shelters are few, casitas may open such areas to colonization by large lobsters while simultaneously aggregating the transient late-stage juveniles, subadults, and adults, making them more accessible to fishing. Tethering experiments have shown that predation on lobsters is lower in and near casitas than in the surrounding seagrass habitat (Eggleston et al. 1990). However, the relative level of protection afforded by casitas depends on the appropriate scaling of shelter, lobster size, and the number of individuals aggregated there (Eggleston et al. 1990; Mintz et al. 1994). There is no compelling evidence that casitas or other large artificial structures used by fisherman enhance the nursery habitat for newly settled juvenile lobsters. Few algal-phase juveniles shelter in large structures, which often attract predators (e.g., snappers, nurse sharks, octopus) of small juvenile lobsters (Mintz et al. 1994), possibly offsetting their value as shelter to these already vulnerable lobster stages. Also, the impact of large artificial structures on the vital rates of subadult or adult lobsters is likely to be less dramatic than what would be manifest if shelter enhancement were to target smaller life stages where

© 1997 NRC Canada

mortality and growth are much greater. Further research on the consequences of the deployment of artificial reefs and structures by fishermen on the growth, reproductive potential, and regional population redistribution of lobsters will provide a more complete picture of their effectiveness in truly enhancing fishery potential (Briones et al. 1994).

We attribute the effectiveness of our artificial shelters in increasing local recruitment of postalgal lobsters to several factors. The small, isolated shelters fulfill the specialized refuge needs of the juveniles most vulnerable to predation and they mimic natural shelters in size and spatial distribution. Scattering the isolated shelters throughout the macroalgal settlement habitat made them easily accessible to lobsters seeking refuge, but their small size and spacing did not make them attractive to large piscine predators. We rarely saw large fish around these small, discrete structures. The artificial shelters also provide refuge as effective against predation as natural structures such as large sponges and seawhips (Childress and Herrnkind 1994). The artificial structures we used are stable, easily deployed, quickly colonized, and camouflaged by epibionts, and they can be used by different cohorts of juvenile lobsters for many years. We have found postalgal juvenile lobsters occupying shelters set out 5 years earlier. However, like natural shelters, the artificial structures we deployed also house other animals that are probable shelter competitors or predators of algal-phase lobsters. Among the former are small stone crabs (*Menippe mercenaria*) and spider crabs (*Mithrax* spp.). Potential predators, as well as den competitors, include toadfish (*Opsanus tau*), swimming crabs (*Portunus* spp.), and octopus. This suite of creatures occurs naturally among large sponges and solution holes but may benefit also from the addition of artificial structures, as has been demonstrated for stone crabs (Beck 1995).

Because of the apparent effectiveness of our small artificial shelters, it is tempting to promote their widespread use, but we urge caution. In other areas of the Caribbean the character of the habitat in shallow inshore areas and abundance of postlarvae differ remarkably from that in the Florida Keys. Thus, it would not be surprising to find that the particular dynamics of shelter availablity and settlement limitation with respect to spiny lobster recruitment may operate differently in other regions. The potential negative impacts of large-scale deployment of artificial structures on the dynamics of the surrounding nursery community are also of ecological and practical interest. The influence of artificial structures or reefs on nearby natural communities is not well known and is likely to be strongly dependent on the species colonizing the structure and the makeup of the surrounding habitat (Davis et al. 1982; Ambrose and Anderson 1990). By virtue of their abundance, mobility, and predatory nature, spiny lobsters potentially can depress the abundance of epifaunal and infaunal macroinvertebrates and alter benthic communities (Robles et al. 1990). Increasing the accessibility of certain areas, like seagrass meadows, to greater numbers of lobsters by augmenting shelter availability may alter seagrass food web dynamics. If adding artificial structures to an area results in increased densities of lobsters, the potential effects on the surrounding community are simultaneously top-down (e.g., increasing predation on benthic macroinvertebrates) and bottom-up (e.g., decreased availability of lobsters for higher trophic level predators, excluding humans). The possible repercussions of habitat-enhancement efforts on community structure and function remain untested and should be a primary concern for fishery biologists and resource managers.

Acknowledgments

We thank Florida Sea Grant for their continued support of our research on spiny lobster recruitment ecology (R/LR-B-26, R/LR-B-30). The Earthwatch Foundation provided additional funding for this project, and the accommodations offered by the Keys Marine Laboratory are appreciated. We are grateful to those who assisted us in the field, especially K. Smith, R. Raney-Butler, and a dedicated group of Earthwatch volunteers. Romuald Lipcius, Jane Lubchenco, Jeffery Polovina, Anne Thistle, Teresa Turner, and two anonymous reviewers read earlier versions of the manuscript and we thank them for their helpful comments.

References

Addison, J.T., and Bannister, R.C.A. 1994. Re-stocking and enhancement of clawed lobster stocks: a review. Crustaceana, **67**: 131–155.

Ambrose, R.F., and Anderson, T.W. 1990. The influence of an artificial reef on the surrounding infaunal community. Mar. Biol. (Berlin), **107**: 41–52.

Andree, S.W. 1981. Locomotory activity patterns and food items of benthic postlarval spiny lobsters, *Panulirus argus*. Master's thesis, Florida State University, Tallahassee, Fla.

Beck, M.W. 1995. Size-specific shelter limitation in stone crabs: a test of the demographic bottleneck hypothesis. Ecology, **76**: 968–980.

Bohnsack, J.A. 1989. Are high densities of fishes at artificial reefs the result of habitat limitation or behavioral preference? Bull. Mar. Sci. **44**: 631–645.

Bohnsack, J.A. 1991. Ecology of artificial reef habitats and fishes. *In* Artificial habitats for marine and freshwater fisheries. *Edited by* W. Seaman and L. Sprague. Academic Press, New York. pp. 61–107.

Bohnsack, J.A., and Sutherland, D.L. 1985. Artificial reef research: a review with recommendations for future priorities. Bull. Mar. Sci. **37**: 11–39.

Briones, P., Lozano, E., and Eggleston, D.B. 1994. The use of artificial shelters (casitas) in research and harvesting of Caribbean spiny lobsters in Mexico. *In* Spiny lobster management. *Edited by* B.F. Phillips, J.S. Cobb, and J. Kittaka. Blackwell Scientific Press, Oxford. pp. 340–362.

Butler, M.J., IV, and Herrnkind, W.F. 1991. The effect of benthic microhabitat cues on the metamorphosis of spiny lobster, *Panulirus argus*, postlarvae. J. Crustacean Biol. **11**: 23–28.

Butler, M.J., IV, Hunt, J.H., Herrnkind, W.F., Childress, M.J., Bertelsen, R., Sharp, W., Mathews, T., Field, J.M., and Marshall, H.G. 1995. Cascading disturbances in Florida Bay, USA: cyanobacteria blooms, sponge mortality, and implications for juvenile spiny lobsters, *Panulirus argus*. Mar. Ecol. Prog. Ser. **129**: 119–125.

Butler, M.J., IV, Herrnkind, W.F., Hunt, J.H., and Bertelsen, R. 1997. Factors affecting the recruitment of juvenile Caribbean spiny lobsters dwelling in macroalgae. Bull. Mar. Sci. In press.

Caddy, J.F. 1986. Modelling stock–recruitment processes in Crustacea: some practical and theoretical perspectives. Can. J. Fish. Aquat. Sci. **43**: 2330–2344.

Caputi, N., and Brown, R.S. 1993. The effect of environment on puerulus settlement of the western rock lobster (*Panulirus cygus*) in Western Australia. Fish. Oceanogr. **2**: 1–10.

© 1997 NRC Canada

Childress, M., and Herrnkind, W. 1994. The behavior of juvenile Caribbean spiny lobsters in Florida Bay: seasonality, ontogeny, and sociality. Bull. Mar. Sci. **54**: 819–827.

Cruz, R., and Phillips, B.F. 1994. The artificial shelters (pesqueros) used for the spiny lobster (*Panulirus argus*) fisheries in Cuba. *In* Spiny lobster management. *Edited by* B.F. Phillips, J.S. Cobb, and J. Kittaka. Blackwell Scientific Press, Oxford. pp. 323–339.

Cruz, R., Brito, R., Diaz, E., and Lalana, R. 1986. Ecología de la langosta (*Panulirus argus*) al SE de la isla de la Juventad. I. Colonización de arrecifes. Rev. Invest. Mar. **8**: 3–17.

Davis, G.E. 1985. Artificial structures to mitigate marina construction impacts on spiny lobster, *Panulirus argus*. Bull. Mar. Sci. **37**: 151–156.

Davis, N., Van Blaricom, G.R., and Dayton, P.K. 1982. Man-made structures on marine sediments: effects on adjacent benthic communities. Mar. Biol. (Berlin), **70**: 295–303.

Eggleston, D.B., and Armstrong, D.A. 1995. Pre- and postsettlement determinants of estuarine Dungeness crab recruitment. Ecol. Monogr. **65**: 193–216.

Eggleston, D., Lipcius, R., Miller, D., and Coba-Centina, L. 1990. Shelter scaling regulates survival of juvenile spiny lobster, *Panulirus argus*. Mar. Ecol. Prog. Ser. **62**: 79–88.

Fairweather, P.G. 1988. Predation can increase variability in the abundance of prey on seashores. Oikos, **53**: 87–92.

Fairweather, P.G. 1991. Implications of supply side ecology for environmental assessment and management. Trends Ecol. Evol. **6**: 60–63.

Field, J.M., and Butler, M.J. IV. 1994. The influence of temperature, salinity, and postlarval transport on the distribution of juvenile spiny lobsters, *Panulirus argus* (Latreille, 1804) in Florida Bay. Crustaceana, **67**: 26–45.

Fogarty, M.J., Sissenwine, M.P., and Cohen, E.B. 1991. Recruitment variability and the dynamics of exploited marine populations. Trends Ecol. Evol. **6**: 241–246.

Forcucci, D., Butler, M.J., IV, and Hunt, J.H. 1994. Population dynamics of juvenile Caribbean spiny lobster, *Panulirus argus*, in Florida Bay, FL. Bull. Mar. Sci. **54**: 805–818.

Fritz, E.S., Crowder, L.B., and Francis, R.C. 1990. The National Oceanic and Atmospheric Administration plan for recruitment fisheries oceanography research. Fisheries, **15**: 25–31.

Hacker, S.D., and Steneck, R.S. 1990. Habitat architecture and the abundance and body-size-dependent habitat selection of a phytal amphipod. Ecology, **71**: 2269–2285.

Harper, D.E. 1991. Trends in the spiny lobster commercial fishery of Florida, 1960–1990. Natl. Mar. Fish. Ser. Rep. No. MIA-91/92-01.

Heatwole, D.W., Hunt, J.H., and Blonder, B.I. 1992. Offshore recruitment of postlarval spiny lobster, *Panulirus argus*, at Looe Key reef, Florida. Proc. Annu. Gulf Caribb. Fish. Inst. **40**: 429–433.

Herrnkind, W.F. 1980. Movement patterns in palinurid lobsters. *In* The biology and management of lobsters. Vol. I. Physiology and behavior. *Edited by* J.S. Cobb and B.F. Phillips. Academic Press, New York. pp. 349–407.

Herrnkind, W.F. 1983. Movement patterns and orientation of Crustacea. *In* Biology of Crustacea. Vol. 5. Behavior and ecology of Crustacea. *Edited by* F. Vernberg and W. Vernberg. Academic Press, New York. pp. 41–105.

Herrnkind, W.F., and Butler, M.J., IV. 1986. Factors regulating postlarval settlement and juvenile microhabitat use by spiny lobsters, *Panulirus argus*. Mar. Ecol. Prog. Ser. **34**: 23–30.

Herrnkind, W.F., and Butler, M.J., IV. 1994. Settlement of spiny lobsters, *Panulirus argus* in Florida: pattern without predictability. Crustaceana, **67**: 46–64.

Herrnkind, W.F., Butler, M.J., IV, and Tankersley, R.A. 1988. The effects of siltation on recruitment of spiny lobsters, *Panulirus argus*. Fish. Bull. **86**: 331–338.

Herrnkind, W.F., Jernakoff, P., and Butler, M.J., IV. 1994. Puerulus and post-puerulus ecology. *In* Spiny lobster management. *Edited by* B.F. Phillips, J.S. Cobb, and J. Kittaka. Blackwell Scientific Press, Oxford. pp. 213–229.

Hughes, T.P. 1990. Recruitment limitation, mortality, and population regulation in open systems: a case study. Ecology, **71**: 12–20.

Hunt, J.H. 1994. Status of the fishery for *Panulirus argus* in Florida. *In* Spiny lobster management. *Edited by* B.F. Phillips, J.S. Cobb, and J. Kittaka. Blackwell Scientific Press, Oxford. pp. 158–168.

Keough, M.J., and Downes, B.J. 1982. Recruitment of marine invertebrates: the role of active larval choices and early mortality. Oecologia, **54**: 348–352.

Kittaka, J. 1994. Larval rearing. *In* Spiny lobster management. *Edited by* B.F. Phillips, J.S. Cobb, and J. Kittaka. Blackwell Scientific Press, Oxford. pp. 402–423.

Lellis, W.A., and Russell, J.A. 1990. Effect of temperature on survival, growth and feed intake of postlarval spiny lobsters, *Panulirus argus*. Aquaculture, **90**: 1–9.

Lewin, R. 1986. Supply-side ecology. Science (Washington, D.C.), **234**: 25–27.

Lewis, J.B., Moore, H.B., and Babis, W. 1952. The postlarval stages of the spiny lobster *Panulirus argus*. Bull. Mar. Sci. **2**: 324–337.

Little, E.J., Jr. 1977. Observations on recruitment of postlarval spiny lobsters, *Panulirus argus*, to the south Florida coast. Fla. Mar. Res. Publ. No. 29.

Little, E.J., Jr., and Milano, G.R. 1980. Techniques to monitor recruitment of postlarval spiny lobsters, *Panulirus argus* to the Florida Keys. Fla. Mar. Res. Publ. No. 37.

Lyons, W.G. 1980. Possible sources of Florida's spiny lobster population. Proc. Gulf Caribb. Fish. Inst. **33**: 253–266.

Marx, J.M. 1986. Recruitment and settlement of spiny lobster pueruli in south Florida. Can. J. Fish. Aquat. Sci. **43**: 2221–2227.

Marx, J.M., and Herrnkind, W.F. 1985. Macroalgae (Rhodophyta: *Laurencia* spp.) as habitat for young juvenile spiny lobsters, *Panulirus argus*. Bull. Mar. Sci. **36**: 423–431.

Miller, T.J., Crowder, L.B., Rice, J.A., and Marshall, E.A. 1988. Larval size and recruitment mechanisms in fishes: toward a conceptual framework. Can. J. Fish. Aquat. Sci. **45**: 1657–1670.

Mintz, J.D., Lipcius, R.N., Eggleston, D.B., and Seebo, M.S. 1994. Survival of juvenile Caribbean spiny lobster: effects of shelter size, geographic location, and conspecific abundance. Mar. Ecol. Prog. Ser. **112**: 255–266.

Moriyasu, M., Landsburg, W., and Conan, G.V. 1994. Sphyrion tag shedding and tag induced mortality of American lobster, *Homarus americanus* H. Milne Edwards, 1837 (Decapoda, Nephropidae). Crustaceana, **68**: 184–192.

National Fisherman. 1986. Artificial habitats could hike crab and lobster catches. National Fisherman December 1986. pp. 10–12 and 64.

Norman, C.P., Yamakawa, H., and Yoshimura, T. 1994. Habitat selection, growth rate, and density of juvenile *Panulirus japonicus* (Von Siebold, 1824) (Decapoda, Panlinuridae) at Banda, Chiba Prefecture, Japan. Crustaceana, **66**: 366–383.

Parrish, F.A., and Polovina, J.J. 1994. Habitat thresholds and bottlenecks in production of the spiny lobster (*Panulirus marginatus*) in the northwestern Hawaiian Islands. Bull. Mar. Sci. **54**: 151–163.

Phillips, B.F. 1986. Prediction of commercial catches of the western rock lobster *Panulirus cygnus*. Can. J. Fish. Aquat. Sci. **43**: 2126–2130.

Phillips, B.F., Palmer, M.J., Cruz, R., and Trendall, J.T. 1992. Estimating growth of the spring lobsters *Panulirus cygnus*, *P. argus*, and *P. ornatus*. Aust. J. Mar. Freshwater Res. **43**: 1177–1188.

© 1997 NRC Canada

Polovina, J.J. 1991. Fisheries applications and biological impacts of artificial habitats. *In* Artificial habitats for marine and freshwater fisheries. *Edited by* W. Seaman and L. Sprague. Academic Press, New York. pp. 154–176.

Possingham, H.P., and Roughgarden, J. 1990. Spatial population dynamics of a marine organism with a complex life cycle. Ecology, **71**: 973–985.

Richards, W.J., and Bohnsack, J.A. 1987. The Caribbean Sea: a large marine ecosystem in crisis. *In* Large marine ecosystems: pattern, processes, and yields. *Edited by* K. Sherman, L.M. Anderson, and B.D. Gold. American Association for the Advancement of Science, Washington, D.C. pp. 44–53.

Robles, C., Sweetman, D., and Eminike, J. 1990. Lobster predation on mussels: shore-level differences in prey vulnerability and predator preference. Ecology, **71**: 1564–1577.

Rosenberg, A.A., Fogarty, M.J., Sissenwine, M.P., Beddington, J.R., and Shepard, J.G. 1993. Achieving sustainable use of renewable resources. Science (Washington, D.C.), **262**: 828–829.

Rothschild, B.J. 1986. Dynamics of marine fish populations. Harvard University Press, Cambridge, Mass.

Rothschild, B.J., Ault, J.S., and Smith, S.G. 1996. A systems science approach to fisheries stock assessment and management. *In* Stock assessment: quantitative methods and applications for small-scale fisheries. *Edited by* V.F. Gallucci, S. Saila, D. Gustafson, and B.J. Rothschild. Lewis Publishers, CRC Press, New York. pp. 473–492.

Roughgarden, J., Gaines, S., and Possingham, H. 1988. Recruitment dynamics in complex life cycles. Science (Washington, D.C.), **241**: 1460–1466.

Sale, P.F. 1990. Recruitment of marine species: is the bandwagon rolling in the right direction? Trends Ecol. Evol. **5**: 25–27.

Scarratt, D.J. 1968. An artificial reef for lobsters (*Homarus americanus*). J. Fish. Res. Board Can. **25**: 2683–2690.

Silberman, J.D., and Walsh, P.J. 1994. Population genetics of the spiny lobster *Panulirus argus*. Bull. Mar. Sci. **54**: 1084.

Sissenwine, M.P. 1984. Why do fish populations vary? *In* Exploitation of marine communities. *Edited by* R.M. May. Springer-Verlag, New York. pp. 59–94.

Smith, K.N., and Herrnkind, W.F. 1992. Predation on juvenile spiny lobsters, *Panulirus argus*: influence of size, shelter, and activity period. J. Exp. Mar. Biol. Ecol. **157**: 3–18.

Spanier, E., and Zimmer-Faust, R.K. 1988. Some physical properties of shelter that influence den preferences in spiny lobsters. J. Exp. Mar. Biol. Ecol. **121**: 137–149.

Steger, R. 1987. Effects of refuges and recruitment on gonodactylid stomatopods, a guild of mobile prey. Ecology, **68**: 1520–1533.

Stone, R.B., McGurrin, J.M., Sprague, L.M., and Seaman, W., Jr. 1991. Artificial habitats of the world: synopsis and major trends. *In* Artificial habitats for marine and freshwater fisheries. *Edited by* W. Seaman and L. Sprague. Academic Press, New York. pp. 31–60.

Tangley, L. 1987. Enhancing coastal production: do artificial shelters help or hurt Mexico's valuable spiny lobster fishery? BioScience, **37**: 309–312.

Underwood, A.J., and Fairweather, P.G. 1989. Supply side ecology and benthic marine assemblages. Trends Ecol. Evol. **4**: 16–19.

Victor, B.C. 1986. Larval settlement and juvenile mortality in a recruitment-limited coral reef fish population. Ecol. Monogr. **56**: 145–160.

Wahle, R., and Steneck, R.S. 1991. Recruitment habitats and nursery grounds of the American lobster *Homarus americanus*: a demographic bottleneck? Mar. Ecol. Prog. Ser. **69**: 231–243.

Yoshimura, T., and Yamakawa, H. 1988. Ecological investigations of settled puerulus and juvenile stages of the Japanese spiny lobster, *Panulirus japonicus*, at Kominato, Japan. J. Crustacean Biol. **8**: 524–531.

Yoshimura, T., Yamakawa, H., and Norman, C.P. 1994. Comparison of hole and seaweed habitats of post-settled pueruli and early benthic juvenile lobsters, *Panulirus japonicus* (Von Siebold, 1824). Crustaceana, **66**: 356–365.

Zieman, J.C., Fourqurean, J.W., and Iversen, R.L. 1989. Distribution, abundance, and productivity of seagrasses and macroalgae in Florida Bay. Bull. Mar. Sci. **44**: 292–311.

© 1997 NRC Canada

A Bayesian observation error model to predict cyanobacterial biovolume from spring total phosphorus in Lake Mendota, Wisconsin

C.A. Stow, S.R. Carpenter, and R.C. Lathrop

Abstract: We developed a logistic model for predicting summer blue-green biovolume from mean (log metric) spring total phosphorus concentration in Lake Mendota, Wisconsin. The model incorporates uncertainty in the sample estimates of the "true" mean total phosphorus values. We used Bayes Theorem to assess model parameters and predictive uncertainty from 19 years of data. When compared with a "naive" model that does not accommodate phosphorus uncertainty, the observation error model has a higher parameter variance, but lower prediction uncertainty. Lower prediction uncertainty occurs because some of the noise in the data is resolved as phosphorus uncertainty, thus reducing the variance of the model disturbance term. The observation error model results in less stringent phosphorus targets to meet acceptable blue-green levels than does the naive model because of this lower prediction uncertainty.

Résumé : Nous avons élaboré un modèle logistique pour prévoir le biovolume d'algues bleu-vert en été à partir de la concentration moyenne (log métrique) de phosphore total au printemps dans le lac Mendota, au Wisconsin. Le modèle tient compte de l'incertitude dans les estimations, basées sur les échantillons, des valeurs moyennes "réelles" du phosphore total. Nous utilisons le théorème de Bayes pour évaluer les paramètres du modèle et l'incertitude de prévision pour 19 années de données. Lorsqu'on compare avec un modèle "naïf" qui ne prend pas en considération l'incertitude liée au phosphore, le modèle tenant compte de l'erreur d'observation présente une variance plus élevée des paramètres, mais une incertitude de prévision plus faible. L'incertitude de prévision plus faible survient parce qu'une partie du bruit dans les données est traitée comme une incertitude liée au phosphore, réduisant ainsi la variance de du terme de perturbation du modèle. Le modèle tenant compte de l'erreur d'observation entraîne des objectifs moins rigoureux en ce qui a trait à la concentration de phosphore pour obtenir des concentrations d'algues bleu-vert acceptables que ne le fait le modèle naïf, en raisons de cette incertitude de prévision plus faible.
[Traduit par la Rédaction]

Introduction

Addressing cultural eutrophication was one of the first tasks of the newly formed U.S. Environmental Protection Agency in the 1970s. At that time, considerable effort was spent assessing the trophic status of lakes in the United States, identifying the primary limiting nutrient, and imposing controls on point-source effluents. Marked improvements occurred, in some lakes, in response to these efforts (Larson 1996), and research efforts have moved into other, often more subtle, environmental problems. However, obnoxious algal blooms are still a problem in numerous lakes in developed watersheds where non-point-source nutrient loads from agricultural and urban runoff are significant (National Research Council 1992).

Models relating nutrients to lake productivity developed rapidly during the 1970s and early 1980s. These models were typically fit to a cross section of lakes and have been commonly referred to as "input–output," "black-box," or "Vollenweider" models (Reckhow and Chapra 1983).

Since the early 1980s, there has been an increasing recognition that cross-sectional models, while valuable for exploring broad patterns over many lakes, have limited predictive value when applied to individual lakes. Large cross-sectional data bases provide a wide range of values for predictor variables so that model parameters are well determined, but the range of the predictors in a single lake is often so limited in comparison with the cross-sectional data base that the predictive precision of the model is insufficient to be helpful for management decisions. Reckhow and Chapra (1983) examined a cross-sectional phosphorus model, fit to data from 290 natural and 433 artificial lakes in Canada, Europe, and the United States, and found systematic biases in some areas of the sample space and prediction intervals with an extremely wide range.

One approach to improve prediction in individual lakes has been to group lakes with some commonality, such as geographic region, in the cross-sectional data set (Reckhow 1987; Trimbee and Prepas 1987). Another tactic, still using cross-sectional data, is to fit parameters to the individual lakes using a random coefficients model (Reckhow 1993).

Our approach was to calibrate a model to many years of data from a single lake. This approach is limited in many

Received January 8, 1996. Accepted July 15, 1996.
J13246

C.A. Stow[1] **and S.R. Carpenter.** University of Wisconsin, Center for Limnology, 680 North Park Street, Madison, WI 53706, U.S.A.
R.C. Lathrop. Bureau of Research, Wisconsin Department of Natural Resources, 1350 Femrite Drive, Monona, WI 53716, U.S.A.

[1] Present address: Nicholas School of the Environment, Duke University, Box 90328, Durham, NC 27708-0328, U.S.A.

© 1997 NRC Canada

instances by the absence of sufficient data from a given lake, particularly if modeling is based on annual phenomena. Even in well-studied lakes, current data bases for trophic characteristics probably contain only 20–25 years of information, resulting in relatively few observations. Consequently, model parameters may be poorly determined and model uncertainty may be high. However, systematic biases will be removed from models calibrated to individual lakes, and predictive precision may be adequate for useful management inference. For most management decisions, parameter values are not of interest anyway; predictions are the quantity on which decisions are based. Additionally, our approach incorporates the model uncertainty into decision-making. Uncertainty analysis often consists of a perfunctory confidence interval calculation without formal inclusion of uncertainty into the decision-making process.

Our goal in this study was to develop a model to assist in setting target total phosphorus concentrations for Lake Mendota, a 3985-ha lake in a highly developed agricultural and urban watershed in southern Wisconsin. Despite the removal of all point-source effluents and an attempt at biological control, Lake Mendota still experiences extreme blue-green algal conditions and a generally decreased water clarity resulting from high non-point-source phosphorus input (Lathrop 1992). This exercise was part of a larger non-point-source pollution abatement project initiated by the Wisconsin Department of Natural Resources.

We modeled summer blue-green algal levels as a function of spring total phosphorus concentrations (Dillon and Rigler 1974). Summer algal densities in Lake Mendota are related to phosphorus inputs resulting from spring rains and snowmelt that combine to flush phosphorus from agricultural land not yet covered by crops (Soranno et al. 1996). Spring phosphorus concentrations also reflect residual phosphorus levels from the preceding fall and winter (Lathrop 1992). Thus, spring phosphorus concentrations are an integrated measure of the lake nutrient status from the previous year and those nutrients added during the spring runoff period before stratification. Modeling summer algae as a function of spring phosphorus also lends a cause and effect interpretation to our model, differing from most previous models, which have related mean algal concentrations to concurrent rather than prior phosphorus levels. In models with high temporal aggregation, functionally relating concurrent mean algal and phosphorus concentrations implicitly relates algal levels occurring early in the aggregation period to subsequent phosphorus levels, considerably reducing the cause and effect sense of the model.

We used a logistic function to relate phosphorus concentrations to blue-green concentrations. Sigmoid relationships between total phosphorus and measures of algal concentration have been demonstrated in cross-sectional analyses (McCauley et al. 1989; Watson et al. 1992; Chow-Fraser et al. 1994) but have not been previously used to describe the association between total phosphorus and blue-green algal concentrations in a single lake.

This analysis differs further from earlier studies by incorporating individual blue-green observations in the model estimation, rather than summary statistics such as the sample mean. Extreme conditions are often more important to lake managers and the general public than average conditions, and linking the probability of extreme conditions to mean conditions is a useful product of model development (Walker 1985).

Statistical problems also arise by using sample means as the dependent variable in a regression analysis because sample means are unlikely to have a common variance, particularly if the sample sizes were very different. Unequal variances can be accommodated with a weighted regression, but in practice this is rarely done. Additionally, log transforming sample means introduces slight biases depending on the sample size and underlying variance (Stow et al. 1995). Ignoring these statistical quirks may be inescapable; summary data are often the only measures that survive from data collection to public access, and the practical consequences may be minor. However, if the necessary data are available, these problems can be avoided.

Another difference between this analysis and prior studies is the incorporation of predictor variable uncertainty into the model. Problems associated with predictor uncertainty are inherent in many published cross-sectional lake models based on mean estimates of the predictor variables. Sample statistics such as means are uncertain estimates of unknown "true" values. Using sample estimates as surrogates for the true values results in biases in parameter estimators (Fuller 1987). In nonlinear models the degree of parameter bias can vary considerably with sample and parameter space, potentially making the model unreliable for some lakes in the calibration data set (Stow and Reckhow 1996). A common justification for ignoring predictor uncertainty is the belief that, for prediction purposes, incorporating predictor uncertainty is unnecessary. However, Fuller (1987) pointed out that this is true only under specific conditions. Models incorporating predictor uncertainty are commonly referred to as "errors-in-variables" or "measurement error" models (Fuller 1987; Caroll et al. 1995). We have used "observation error" as a more general terminology to incorporate sampling error, inherent in sample mean data, in addition to measurement error.

A difficulty that occurs in observation error models is that each predictor variable observation results in an "incidental" parameter (Zellner 1971). Values of the incidental parameters are not typically of interest, but the presence of many incidental parameters causes model calibration to become a high-dimensional optimization problem. We estimated model parameters with Bayes Theorem and treated the incidental parameters as nuisance parameters, integrating them out of the posterior parameter distribution. We compared the observation error model predictions and parameter estimates with predictions and parameter estimates from an analogous "naive" model that ignores total phosphorus uncertainty.

Methods

The data in this analysis are from the 19-year period 1976–1994. This period encompasses extremes in phosphorus loading events, including the drought years from 1987 to 1988 and heavy rains and flooding in 1993. All sampling was conducted at the deepest spot in Lake Mendota. Spring total phosphorus samples were composited from subsamples collected at 0 and 4 m during the period between ice-out and the onset of stratification, typically the end of March to early May. We used samples collected from July–September to represent summer blue-green algal densities. Densities were

© 1997 NRC Canada

determined on 0- to 2-m composite samples from 1976 to 1987, 0- to 10-m composites from 1988 to 1989, and 0- to 8-m composites beginning in 1990. Phytoplankton samples were identified and counted using an inverted microscope (Utermöhl) technique, and individual species biovolumes were estimated from cell measurement data on each sample. Detailed descriptions of sample collection and analysis protocol for phosphorus and phytoplankton are described in Lathrop and Carpenter (1992).

Using nonlinear least squares on sample mean data, we explored the fit of two alternative models, a simple linear model and a logistic function of the form

$$BG = \frac{\alpha}{1 + \exp[\beta_0 + \beta_1 TP]}$$

where BG was the mean summer blue-green biovolume (microlitres per litre), TP was the mean spring total phosphorus concentration (milligrams per litre), and α, β_0, and β_1 were unknown parameters. In the exploratory analysis, we recognized that the three parameters in the logistic function were individually poorly determined and highly covarying and that optima for α and β_0 were approximately equal. To reduce the model dimension, we replaced α with β_0 and created a two-parameter model; the parameters in this model were also poorly determined, and highly correlated, so we further reduced the model dimension by replacing β_1 with $-8 \times \beta_0$, the approximate relationship between optimal values of the two parameters. Based on these exploratory results, we modeled median summer blue-green biovolume (millitres per litre) as a single-parameter logistic function of median spring total phosphorus concentration (milligrams per litre). We estimated β_0 for two models: a "naive" model that ignores the observation error in total phosphorus and an observation error model incorporating the predictor variable error structure. The observation error model had a total of 21 unknown parameters: one logistic parameter, the model variance parameter, and 19 incidental parameters, one associated with each of 19 spring phosphorus conditions. The naive model had a total of two unknowns: one logistic parameter and the variance parameter. We estimated model parameters using Bayes Theorem. Because of the high dimensionality of the problem, we chose a Gauss-Hermite quadrature approximation (Piessens et al. 1983) as opposed to a Monte-Carlo technique such as importance sampling (Geweke 1989; Wolpert 1991; McAllister et al. 1994) to integrate the 19 incidental parameters from the posterior distribution of the observation error model. Model development is detailed in the Appendix.

Results

Exploratory analysis using sample means indicated that all three forms of the logistic model provided a better fit to the data than a simple linear model, with higher R^2 values and comparable or lower mean squared errors (Table 1). Reparameterizing the original three-parameter logistic to two- and one-parameter forms did not affect R^2 values but did decrease the model mean squared error. Reexpressing the original three parameters as two parameters or one did not affect the "fit" of the model, as indicated by identical R^2 values, because the parameters were so highly covarying. However, the mean squared error decreased with fewer parameters because the degrees of freedom increased. Exploratory analysis also revealed that sample means for both summer blue-green biovolume and spring total phosphorus are relatively uncertain estimates (Fig. 1). This uncertainty, particularly in spring total phosphorus, invites a measurement error approach explicitly incorporating uncertainty.

A comparison of expected values for the observation error and naive models indicates that both provide a reasonable fit to the sample mean data (Fig. 2), considering the uncertainty associated with each sample mean (Fig. 1). Differences occur because the logistic function is not monotonic in the parameter β_0, and posterior distributions for β_0 are rather different for the two models (Fig. 3). In effect, the observation error model assumes that the marginal variability of total phosphorus is less than indicated by the sample mean phosphorus values. Thus, the independent variable axis in Fig. 2 is compressed, leading to a steeper relationship between blue-green biovolume and total phosphorus. In addition to different optima and mean values, the variance of the posterior marginal distribution for β_0 in the observation error model is slightly higher than for β_0 in the naive model. A higher variance for β_0 in the observation error model indicates that model uncertainty increases when predictor uncertainty is recognized in the model estimation.

Table 1. Exploratory analysis summary statistics.

Model	Mean squared error	R^2
Simple linear	0.850	0.40
Three-parameter logistic	0.898	0.51
Two-parameter logistic	0.848	0.51
One-parameter logistic	0.806	0.51

However, although β_0 is slightly less certain when predictor uncertainty is considered, the variance of the model disturbance term, σ_{bg}^2 is also considerably less (Fig. 4). When parameter variance and disturbance term variance are combined in the predictive distribution, there is a net decrease in the prediction variance (Fig. 5). Parameter estimation is (falsely) more precise when predictor uncertainty is ignored, but prediction precision is lower. It is interesting to note that although prediction precision differs considerably between the two models, the most likely predictions are fairly similar (Fig. 5).

Discussion

This study further illustrates the limitations of cross-sectional data for making useful management decisions in a particular lake. The Lake Mendota total phosphorus sample means range from about 0.06 to 0.18 $mg \cdot L^{-1}$ and the blue-green sample means range from 0.3 to 8 $\mu L \cdot L^{-1}$. These values contrast with the cross-sectional data analyzed by Watson et al. (1992), which range from about 0.003 to 1.0 $mg \cdot L^{-1}$ for phosphorus and from 0.1 to 100 $mL \cdot L^{-1}$ for algal biovolume. The Mendota data overlap a small subspace of these cross-sectional data, and within this subspace, the cross-sectional data are essentially patternless. Predictions from a model based on these cross-sectional data would be too imprecise to delineate management options for Lake Mendota.

Models can be helpful tools for making decisions. Ultimately, however, most decisions require some fairly arbitrary judgment such as appropriate decision criteria (maximizing expected utility, minimizing maximum risk), acceptable probability levels, in this case acceptable algae levels, or some other value-based standard. Models can provide guidance for choosing such criteria or elucidate an acceptable class of management actions, but subjective input is usually necessary.

© 1997 NRC Canada

Fig. 1. Uncertainty in the sample means as a measure of the true unknown means for summer blue-green biovolume (left panel) and spring total phosphorus concentration (right panel). Uncertainty intervals of ±2 SE on each side of the sample mean are depicted.

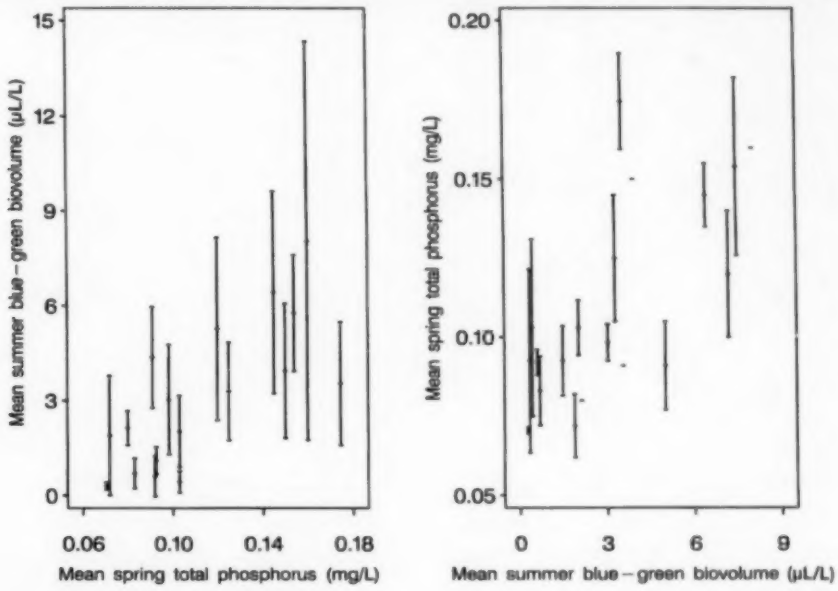

Mean summer blue–green biovolume (µL/L)
Mean spring total phosphorus (mg/L)
Mean spring total phosphorus (mg/L)
Mean summer blue–green biovolume (µL/L)

Fig. 2. Mean model values for the observation error (circles) and naive (squares) models plotted among sample mean observations (represented by the years 1976–1994).

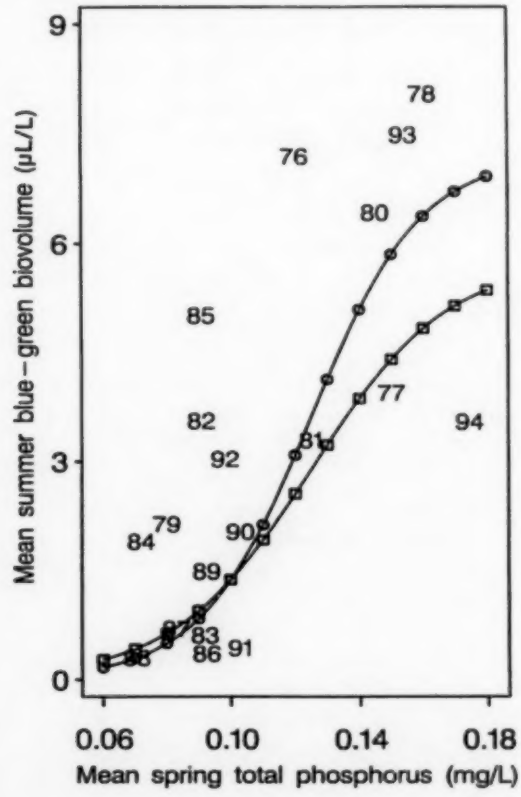


Fig. 3. Comparison of observation error and naive model marginal posterior distributions for parameter β_0.

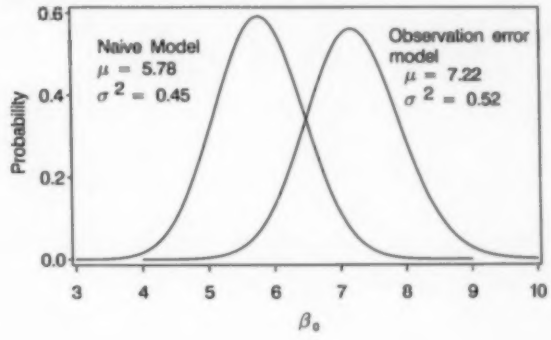
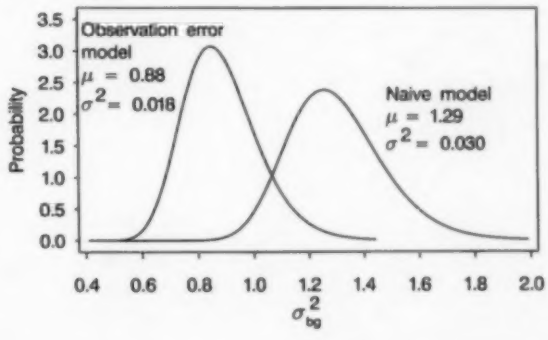
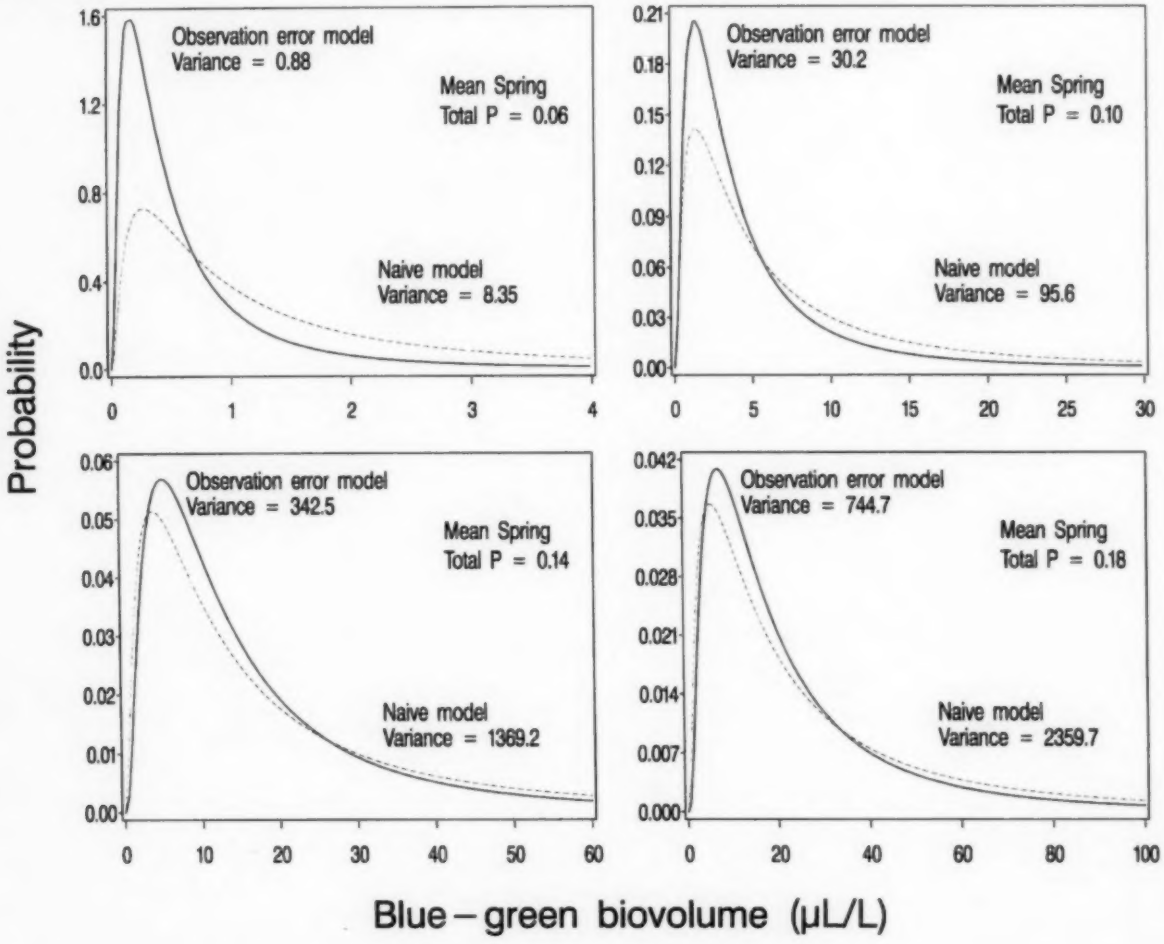


Fig. 4. Comparison of observation error and naive model marginal posterior distributions for the disturbance term variance, σ_{bg}^2.



© 1997 NRC Canada

Fig. 5. Comparison of predictive distributions for the observation error (solid line) and naive (broken line) models, conditional on four mean spring total phosphorus concentrations (milligrams per litre).



Both the naive and observation error models indicate a region of relatively rapid algal response to changes in phosphorus concentration (Fig. 2). Setting target phosphorus levels below this region, less than 0.10 $mg \cdot L^{-1}$, for example, might be a reasonable management goal. Even fairly low phosphorus concentrations include some risk of extreme algal events (Fig. 5). The task for decision makers is establishing a tolerable risk.

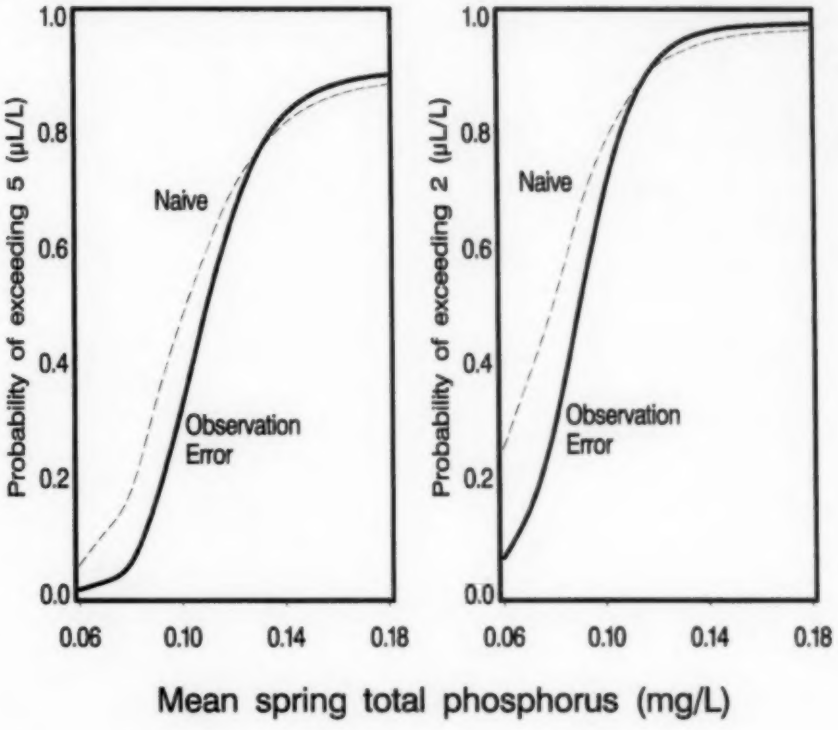
Phosphorus input is not the only factor responsible for nuisance algal conditions. However, non-point-source phosphorus input is one of the few important components that active management can potentially limit. More complex models relating lake productivity to environmental variables in addition to phosphorus may, in principal, produce more precise predictions. However, precise predictions conditioned on specific values of stochastic and essentially uncontrollable factors are minimally useful for management projections. We could have developed a more complex model with more terms to accommodate such things as light, carbon dioxide, N:P ratios, or any of a number of factors that have been suggested to be important (Smith 1986; Shapiro 1990), but each additional variable would introduce at least one more unknown parameter into the model and further increase the dimension of the problem.

"The curse of dimensionality" can be especially troubling in Bayesian analysis because high-dimensional integrals are often involved in model estimation. Reducing the model from a three-parameter logistic function by reparameterizing to a single parameter is a convenient manipulation that does not appreciably affect model performance but greatly reduces the time to perform numerical integrations.

Reduction to a single-parameter logistic function also has considerable heuristic value because the relative uncertainties of the observation error and naive models can be directly compared by comparing the posterior plots of β_0. Uncertainty propagation in higher dimensional nonlinear models can be complex, especially if the parameters covary and the model is not monotonic in the parameters. Encapsulating model uncertainty in a single parameter reveals that accounting for observation error in total phosphorus increases model

© 1997 NRC Canada

Fig. 6. Comparison of estimated probabilities for exceeding two hypothetical target blue-green concentrations using the observation error (solid line) and naive (broken line) models.



uncertainty, as indicated by a higher posterior variance for β_0 (Fig. 3). In other words, if we are less certain of our data, we will be less certain of the location of the curve through the data.

Predictive precision is determined by both the model uncertainty, as indicated by the posterior variance of β_0 (Fig. 3), and the variance of the model disturbance, σ_{bg}^2 (Fig. 4). These two variance components propagate through the model structure and produce the predictive distributions in Fig. 5. Variance propagation is not analytically straightforward as it is in a simple linear model, so these predictive distributions must be estimated numerically. The net result, in this example, is that the prediction uncertainty is lower in the observation error model, even though the parameter uncertainty is slightly higher. In essence, some of the noise in the data is absorbed as uncertainty in total phosphorus, substantially lowering σ_{bg}^2, the noise in blue-green algae. Total phosphorus uncertainty makes us less able to precisely determine β_0, but predictions conditioned on specific total phosphorus values are more precise. While the posterior distribution of β_0 has a lower variance in the naive model, this apparent precision is deceiving because the model is based on the incorrect assumption that total phosphorus is known exactly.

Prediction precision differences can have important management implications. Estimated probabilities of exceeding target blue-green concentrations are higher in the naive model by four-to five-fold at lower phosphorus concentrations (Fig. 6). At higher phosphorus concentrations, predictions become more similar; both models predict that target blue-green levels will be exceeded if phosphorus levels are high. However, the region of interest for management is at the lower phosphorus levels where the large differences occur.

For developing phosphorus concentration standards to meet targeted blue-green levels, both the observation error and naive models are conservative, as they predict higher blue-green values than have actually been observed, particularly at high phosphorus levels (Fig. 5). The highest individual blue-green observations in this analysis were about 20 $\mu L \cdot L^{-1}$, but both models estimate values in excess of 20 to be probable at spring phosphorus levels of 0.14 and 0.18 $mg \cdot L^{-1}$. This apparent overprediction arises from the propagation of uncertainty; because model parameters are relatively uncertain (Fig. 3), extremely high blue-green concentrations cannot be ruled out. The naive model overpredicts more than the observation error model because, in the naive model, all noise in the data is resolved as uncertainty in blue-green concentration. In the observation error model, some noise is resolved as uncertainty in total phosphorus concentration. If the observation error model is closer to "truth" than the naive model, then it will provide a more proper translation of data variability into management actions that formally incorporate uncertainty.

Prairie et al. (1995) have stated that variability is a "curse" that obfuscates empirical relationships. This statement is especially true when empirical relationships are used to justify management decisions where economic stakes are high.

© 1997 NRC Canada

Effective management requires decision makers to confront uncertainty and integrate it into policy (Ludwig et al. 1993). We have developed a model that incorporates phosphorus variability and propagates this variability into blue-green prediction uncertainty. Inclusion of phosphorus variability results in a lower prediction uncertainty and illustrates another reason why considering such variability can be useful.

Acknowledgments

Pat Soranno, Doran Mason, and Tara Stow provided helpful reviews of this manuscript. This work was supported by the A.W. Mellon Foundation, the North Temperate Lakes LTER Program, and the Wisconsin Department of Natural Resources.

References

Caroll, R.J., Ruppert, D., and Stefanski, L.A. 1995. Measurement error in nonlinear models. Chapman and Hall, New York.

Chow-Fraser, P., Trew, D.O., Findlay, D., and Stainton, M. 1994. A test of hypotheses to explain the sigmoidal relationship between total phosphorus and chlorophyll a concentrations in Canadian lakes. Can. J. Fish. Aquat. Sci. **51**: 2052–2065.

Dillon, P.J., and Rigler, F.H. 1974. The phosphorus–chlorophyll relationship in lakes. Limnol. Oceanogr. **19**: 767–773.

Fuller, W.A. 1987. Measurement error models. John Wiley & Sons, New York.

Geweke, J. 1989. Bayesian inference in econometric models using Monte Carlo integration. Econometrica, **57**: 1317–1339.

IMSL. 1987. User's manual — Math Library. International Mathematics and Statistics Library, Houston, Tex.

Larson, D.W. 1996. Curing the incurable? Am. Sci. **84**: 7–9.

Lathrop, R.C. 1992. Nutrient loadings, lake nutrients, and water clarity. *In* Food web management, a case study of Lake Mendota. *Edited by* J.F. Kitchell. Springer-Verlag, New York. pp. 69–96.

Lathrop, R.C., and Carpenter, S.R. 1992. Phytoplankton and their relationship to nutrients. *In* Food web management, a case study of Lake Mendota. *Edited by* J.F. Kitchell. Springer-Verlag, New York. pp. 97–126.

Ludwig, D., Hilborn, R., and Walters, C. 1993. Uncertainty, resource exploitation, and conservation: lessons from history. Science (Washington, D.C.), **260**: 17, 36.

McAllister, M.K., Pikitch, E.K., Punt, A.E., and Hilborn, R. 1994. A Bayesian approach to stock assessment and harvest decisions using the sampling/importance resampling algorithm. Can. J. Fish. Aquat. Sci. **51**: 2673–2686.

McCauley, E., Downing, J.A., and Watson, S. 1989. Sigmoid relationships between nutrients and chlorophyll among lakes. Can. J. Fish. Aquat. Sci. **46**: 1171–1175.

National Research Council. 1992. Restoration of aquatic ecosystems — science, technology and public policy. National Academies Press, Washington, D.C.

Piessens, R., deDoncker-Kapenga, C., Uberhuber, W., and Kahaner, D.K. 1983. Quadpack. Springer-Verlag, New York.

Prairie, Y.T., Peters, R.H., and Bird, D.F. 1995. Natural variability and the estimation of empirical relationships: a reassessment of regression methods. Can. J. Fish. Aquat. Sci. **52**: 788–798.

Reckhow, K.H. 1987. Empirical models for trophic state in southeastern U.S. lakes and reservoirs. Water Resour. Bull. **24**: 723–734.

Reckhow, K.H. 1993. A random coefficient model for chlorophyll–nutrient relationships in lakes. Ecol. Model. **70**: 35–50.

Reckhow, K.H., and Chapra, S.C. 1983. Engineering approaches for lake management. Vol. 1. Data analysis and empirical modeling. Butterworth Publishers, Boston, Mass.

Shapiro, J. 1990. Current beliefs regarding dominance by blue-greens: the case for the importance of CO2 and pH. Verh. Int. Ver. Limnol. **24**: 38–54.

Smith, V.H. 1986. Light and nutrient effects on the relative biomass of blue-green algae in lake phytoplankton. Can. J. Fish. Aquat. Sci. **43**: 148–153.

Soranno, P.A., Hubler, S.L., Carpenter, S.R., and Lathrop, R.C. 1996. Phosphorus loads to surface waters: a simple model to account for effects of land use. Ecol. Appl. **6**: 865–878.

Stow, C.A., and Reckhow, K.H. 1996. Estimator bias in a lake phosphorus model with observation error. Water Resour. Res. **32**: 165–170.

Stow, C.A., Carpenter, S.R., Amrhein, J.F., Eby, L.A., and Hesselberg, R.J. 1995. Evidence that PCBs are approaching stable concentrations in Lake Michigan fishes. Ecol. Appl. **5**: 248–260.

Trimbee, A.M., and Prepas, E.E. 1987. Evaluation of total phosphorus as a predictor of the relative biomass of blue-green algae with emphasis on Alberta lakes. Can. J. Fish. Aquat. Sci. **44**: 1337–1342.

Walker, W.W. 1985. Statistical bases for mean chlorophyll a criteria. Lake Reservoir Manage. **4**: 57–62.

Watson, S., McCauley, E., and Downing, J.A. 1992. Sigmoid relationships between phosphorus, algal biomass, and algal community structure. Can. J. Fish. Aquat. Sci. **49**: 2605–2610.

Wolpert, R.L. 1991. Monte Carlo integration in Bayesian statistical analysis. Contemp. Math. **115**: 101–116.

Zellner, A. 1971. An introduction to Bayesian inference in econometrics. John Wiley & Sons, Inc., New York.

Appendix

Development of the observation error model

There are $h = 1, 2, \ldots, n$ years, with $n = 19$. Within each year there are $i = 1, 2, \ldots, m_h$ blue-green observations. m_h ranges from 3 to 12. Within each year there are $j = 1, 2, \ldots, k_h$ phosphorus observations. k_h ranges from 1 to 6. The median summer blue-green biovolume is modeled as a logistic function of the median spring total phosphorus concentration:

$$(1) \qquad \exp(\mathrm{LBG}_h) = \frac{\alpha}{1 + \exp[\beta_0 + \beta_1 \exp(\mathrm{LTP}_h)]}$$

where LBG_h = median summer blue-green biovolume in year h, expressed in the log metric, $\exp(\mathrm{LBG}_h)$ = median summer blue-green biovolume, expressed in the natural metric (microlitres per litre), LTP_h = median spring total phosphorus in year h, expressed in the log metric, and $\exp(\mathrm{LTP}_h)$ = median spring total phosphorus, expressed in the natural metric (milligrams per litre). Under the assumption that individual blue-green and total phosphorus observations are lognormally distributed, mean and median values are the same in the log metric.

© 1997 NRC Canada

Individual blue-green observations are assumed lognormally distributed so that:

$$(2) \qquad \log(bg_{hi}) = \log\left(\frac{\alpha}{1+\exp[\beta_0+\beta_1\exp(LTP_h)]}\right)+\varepsilon_{bghi} \qquad \varepsilon_{bg}\sim N(0,\sigma_{bg}^2)$$

where bg_{hi} = ith blue-green observation in year h, εbg_{hi} is the model disturbance term, and σ_{bg}^2 = variance of the disturbance term. This variance is assumed constant among years. Constant variance in the log metric indicates that the variance in the natural metric will increase with increasing blue-green levels.

If we assume that individual total phosphorus observations are lognormally distributed, then the total phosphorus sample mean, in the log metric, is normally distributed about the true mean, in the log metric:

$$\overline{ltp}_h = TP_h + \varepsilon_{tphj} \qquad \varepsilon_{tp}\sim N(0,\sigma_{tp}^2/k_h)$$

where

$$\frac{\sum_{j=1}^{k_h}\log(tp_{hj})}{k_h} = \overline{ltp}_h,$$

$\overline{ltp}_h$ = spring total phosphorus sample mean, in the log metric, in year h and σ_{tp}^2 = variance, in the log metric, of individual spring total phosphorus measurements, and tp_{hj} are the individual total phosphorus measurements in year h. This variance is assumed constant among years.

The likelihood function is then

$$(3) \qquad L(bg_{hi},\ \overline{ltp}_h,\ \sigma_{tp},\ k_h|\alpha,\ \beta_0,\ \beta_1,\ LTP_{1,2,\ldots,n},\ \sigma_{bg}^2) =$$

$$\prod_{h=1}^{n}\frac{1}{\sqrt{2\pi\sigma_{tp}^2/k_h}}e^{\frac{-(\overline{ltp}_h-LTP_h)^2}{2\sigma_{tp}^2/k_h}}\prod_{i=1}^{m_h}\frac{1}{\sqrt{2\pi\sigma_{bg}^2}}e^{\frac{-\left(\log(bg_{hi})-\log\left\{\frac{\alpha}{1+\exp[\beta_0+\beta_1\exp(LTP_h)]}\right\}\right)^2}{2\sigma_{bg}^2}}.$$

If we use a value for σ_{tp}^2 estimated from data, there are $n = 19$ unknown LTP_h parameters, α, β_0, β_1, and σ_{bg}^2 for a total of 23 unknown parameters.

We can drop the constants from eq. 3 resulting in

$$(4) \qquad L(bg_{hi},\ \overline{ltp}_h,\ \sigma_{tp},\ k_h|\alpha,\ \beta_0,\ \beta_1,\ LTP_{1,2,\ldots,n},\ \sigma_{bg}^2)\propto$$

$$\prod_{h=1}^{n}e^{\frac{-(\overline{ltp}_h-LTP_h)^2}{2\sigma_{tp}^2/k_h}}\prod_{i=1}^{m_h}\frac{1}{\sqrt{\sigma_{bg}^2}}e^{\frac{-\left(\log(bg_{hi})-\log\left\{\frac{\alpha}{1+\exp[\beta_0+\beta_1\exp(LTP_h)]}\right\}\right)^2}{2\sigma_{bg}^2}}.$$

Now let

$$\omega_h = \frac{\overline{ltp}_h - LTP_h}{\sqrt{2\sigma_{tp}^2/k_h}}$$

so that

$$(5) \qquad LTP_h = \overline{ltp}_h - \omega_h\sqrt{2\sigma_{tp}^2/k_h}.$$

Substitute eq. 5 into eq. 4 for LTP_h:

$$(6) \qquad L(bg_{hi},\ \overline{ltp}_h,\ \sigma_{tp},\ k_h|\alpha,\ \beta_0,\ \beta_1,\ \omega_{1,2,\ldots,n},\ \sigma_{bg}^2)\propto\prod_{h=1}^{n}e^{-(\omega_h^2)}\prod_{i=1}^{m_h}\frac{1}{\sqrt{\sigma_{bg}^2}}e^{\frac{-(\log(bg_{hi})-\log(y))^2}{2\sigma_{bg}^2}}$$

where

$$y = \frac{\alpha}{1+\exp[\beta_0+\beta_1\exp(\overline{ltp}_h-\omega_h\sqrt{2\sigma_{tp}^2/k_h})]}.$$

Now the 19 unknown LTP_h parameters have been replaced by 19 unknown ω_h parameters.

© 1997 NRC Canada

Using a noninformative Jeffreys' prior:

$$(7)\qquad \pi(\alpha, \beta_0, \beta_1, \omega_{1,2,\ldots,n}, \sigma_{bg}) = \frac{1}{\sigma_{bg}}.$$

The posterior distribution is proportional to the product of eqs. 6 and 7:

$$(8)\qquad \pi(\alpha, \beta_0, \beta_1, \omega_{1,2,\ldots,n}, {\sigma_{bg}}^2 | bg_{hi}, \overline{ltp}_h, {\sigma_{tp}}^2, k_h) \propto \frac{1}{\sigma_{bg}} \prod_{h=1}^{n} e^{-({\omega_h}^2)} \prod_{i=1}^{m_h} \frac{1}{\sqrt{{\sigma_{bg}}^2}} e^{\frac{-(\log(bg_{hi})-\log(y))^2}{2{\sigma_{bg}}^2}}.$$

The model variance term, ${\sigma_{bg}}^2$, can be consolidated:

$$(9)\qquad \pi(\alpha, \beta_0, \beta_1, \omega_{1,2,\ldots,n}, \sigma_{bg} | bg_{hi}, \overline{ltp}_h, {\sigma_{tp}}^2, k_h) \propto {\sigma_{bg}}^{-((\sum_{h=1}^{n} m_h)+1)} \prod_{h=1}^{n} e^{-({\omega_h}^2)} \prod_{i=1}^{m_h} e^{\frac{-(\log(bg_{hi})-\log(y))^2}{2{\sigma_{bg}}^2}}.$$

Exploratory analysis using nonlinear least squares indicated α, β_0, and β_1 to be individually poorly determined and highly covarying, with $\alpha \approx \beta_0$ and $\beta_1 \approx -8 \times \beta_0$ at the optimum. The high parameter variances and covariance among the parameters suggest that reparameterization in terms of a single parameter may not significantly compromise optimality. In fact, nonlinear least squares exploratory analysis indicated that replacing α with β_0 and β_1 with $-8 \times \beta_0$ yielded a decrease in the model mean squared error. In the Bayesian model, replacing α with β_0 and β_1 with $-8 \times \beta_0$ reduces the dimension of the problem by two and results in the following:

$$(10)\qquad \pi(\beta_0, \omega_{1,2,\ldots,n}, \sigma_{bg} | bg_{hi}, \overline{tp}_h, {\sigma_{tp}}^2, k_h) \propto {\sigma_{bg}}^{-((\sum_{h=1}^{n} m_h)+1)} \prod_{h=1}^{n} e^{-({\omega_h}^2)} \prod_{i=1}^{m_h} e^{\frac{-(\log(bg_{hi})-\log(z))^2}{2{\sigma_{bg}}^2}}$$

where

$$z = \frac{\beta_0}{1 + \exp[\beta_0 - 8\beta_0 \exp(l\overline{tp}_h - \omega_h\sqrt{2{\sigma_{tp}}^2/k_h})]}.$$

This is now a problem in 19 unknown nuisance parameters and two parameters of interest. We integrated the incidental parameters from the posterior distribution of the observation error model using a Gauss-Hermite quadrature rule (Piessens et al. 1983). Gauss-Hermite quadrature is a numerical approximation applicable for integrals of the form

$$\int_{-\infty}^{\infty} \exp(-x^2) f(x) dx$$

where $f(x)$ is a limited range of functional forms of x. The function is calculated at a number of specific values of x and the integral is approximated as a weighted sum of $f(x)$ values. We corroborated the accuracy of these approximations by comparing Gauss-Hermite rules for 16 and 20 $f(x)$ evaluations.

Integrating the 19 ω_hs from the posterior yields a two-dimensional posterior distribution for β_0 and σ_{bg}:

$$(11)\qquad \pi(\beta_0, \sigma_{bg} | bg_{hi}, \overline{ltp}_h, {\sigma_{tp}}^2, k_h) \propto {\sigma_{bg}}^{-((\sum_{h=1}^{n} m_h)+1)} \prod_{h=1}^{n} \int_{\omega_h} e^{-({\omega_h}^2)} \prod_{i=1}^{m_h} e^{\frac{-(\log(bg_{hi})-\log(z))^2}{2{\sigma_{bg}}^2}} \partial\omega_h.$$

Marginal posterior densities for β_0 and σ_{bg} (Figs. 3 and 4, respectively) are obtained by integrating eq. 11 with respect to σ_{bg} and β_0, respectively, using IMSL numerical software (IMSL 1987).

Predictive distributions for future blue-green, bg*, at future total phosphorus levels, LTP*, are calculated as the product of eq. 11 and

$$(12)\qquad \pi(bg^* | \beta_0, {\sigma_{bg}}^2, LTP^*) = \frac{1}{\sqrt{2\pi{\sigma_{bg}}^2}} e^{\frac{-\left(\log(bg^*) - \log\left\{\frac{\beta_0}{1+\exp[\beta_0 - 8\beta_0 \exp(LTP^*)]}\right\}\right)^2}{2{\sigma_{bg}}^2}}$$

integrated with respect to β_0 and ${\sigma_{bg}}^2$.

Development of the naive model

Development of the naive model parallels the observation-error model in eq. 2 except that LTP_h is replaced by $\overline{tp}_h$, which is

© 1997 NRC Canada

assumed (incorrectly) to be an observation, without error, of LTP_h. The model is then

$$\log(bg_{hi}) = \log\left(\frac{\alpha}{1+\exp[\beta_0+\beta_1\exp(\overline{tp}_h)]}\right)+\varepsilon_{bghi} \qquad \varepsilon_{bg}\sim N(0,\sigma_{bg}{}^2) \tag{13}$$

and the likelihood function is

$$L(bg_{hi},\overline{tp}_h|\alpha,\beta_0,\beta_1,\sigma_{bg}{}^2)=\prod_{i=1}^{m_h}\frac{1}{\sqrt{2\pi\sigma_{bg}{}^2}}\,e^{\frac{-\left(\log(bg_{hi})-\log\left\{\frac{\alpha}{1+\exp[\beta_0+\beta_1\exp(\overline{tp}_h)]}\right\}\right)^2}{2\sigma_{bg}{}^2}}. \tag{14}$$

Development is now analogous to the observation error model.

© 1997 NRC Canada

PERSPECTIVE

Fishers' individual salmon harvesting rights: an option for Canada's Pacific fisheries

R. Quentin Grafton and Harry W. Nelson

Abstract: Canada's Pacific salmon (*Oncorhynchus*) fisheries are again in a crisis of declining stocks, excess fishing effort, and poor returns. Changes in fisheries management that may help resolve some of these problems include licence buy-backs, individual transferable quotas, additional effort controls, single-gear licensing, and area licensing or combinations thereof. This paper proposes an alternative: fishers' individual salmon harvesting rights, which provide fishers with rights to bid for the privilege of participating in salmon fishing openings. Fishers' individual salmon harvesting rights have the potential to rationalize the salmon fleets and control excessive fishing effort and thus help ensure the sustainability of the resource and viability of the industry.

Résumé : Les pêcheries canadiennes de saumon du Pacifique (*Oncorhynchus*) sont encore une fois en crise. La situation se caractérise par une diminution des stocks, un effort de pêche excessif et un faible rendement. Parmi les changements au niveau de la gestion des pêches qui pourraient aider à résoudre certains de ces problèmes figurent le rachat de licences, l'attribution de contingents transférables individuels, une réglementation additionnelle de l'effort, l'autorisation d'un seul type d'engin et le recours à des permis de zone ou encore, une combinaison de ces différentes mesures. Le présent article propose une autre solution : les droits individuels de récolte du saumon du pêcheur. Cette mesure donne au pêcheur le droit de faire une soumission pour avoir le privilège de participer à l'ouverture de la pêche au saumon. Les droits individuels de récolte du saumon des pêcheurs ont le potentiel de rationaliser les flottes de pêche au saumon et de contrôler l'effort de pêche excessif et ainsi, d'aider à assurer la durabilité de la ressource et la viabilité de l'industrie.
[Traduit par la Rédaction]

Introduction

Foremost among the problems faced by Canada's Department of Fisheries and Oceans (DFO) in managing the salmon (*Oncorhynchus*) stocks of British Columbia is the need to accurately estimate the timing, size, and routes of the salmon runs so as to allow sufficient fish to escape and spawn (see Hatfield Consultants Ltd. (1996) for an up-to-date review of the state of the salmon resources in British Columbia). This is complicated by the fact that fishers may in just a few hours catch an entire salmon run. In this scenario, improper timing of the length of an opening could spell disaster for the future of the industry. To avoid such disasters, fishery managers try to estimate escapement for the net fisheries through preseason forecasts, an evaluation of the number of salmon that escape upstream of commercial fishing, and commercial catch data. This information is used to forecast future salmon runs and set weekly targets for escapement, which are updated regularly. In the troll fishery, which uses poles and lines and is more geographically dispersed, the monitoring of harvests is less precise. However, if the cumulative troll catch differs during the season significantly from amounts specified at the start of the season, regulations are adjusted as necessary (Healey 1993).

These management procedures are complicated by the fact that competition among commercial fishers in the three gear types (troll, gill net, and seine gear) is increasing fishing power every year while reducing the net returns to the fleet. This, coupled with precipitous declines in certain stocks, such as the Fraser sockeye salmon (*Oncorhynchus nerka*) runs (Fraser River Sockeye Public Review Board 1995), and conservation worries over chinook salmon (*Oncorhynchus tshawytscha*) in the Strait of Georgia, has led the industry and the regulator to examine different management regimes.

Management options for the commercial fleet (DFO 1995*a*) include additional effort controls that regulate the inputs that can be applied by fishers, area licensing that restricts fishers to fixed locales for harvesting, single-gear licensing that would prevent vessels from using more than one gear type, separating the seine fleet into two for management purposes, and individual transferable quotas (ITQs) that set limits on individual catches (Christy 1973; Moloney and Pearse 1979). On

Received March 26, 1996. Accepted July 31, 1996.
J13375

R.Q. Grafton. Department of Economics, University of Ottawa, P.O. Box 450, Station A, Ottawa, ON K1N 6N5, Canada.
H.W. Nelson. Faculty of Forestry, University of British Columbia, Vancouver, BC V6T 1Z1, Canada.

© 1997 NRC Canada

Table 1. Number of salmon (thousands) caught by statistical area in British Columbia between 1984 and 1993 (DFO 1993).

Year	1–2W	3–5	6–8	9–10	12–13	14–18	20	21–27	28–29D	Total
1984	3225	5 995	1 527	216	1 685	414	680	4069	932	18 743
1985	2826	7 805	5 025	791	8 950	1169	5214	7175	1939	40 894
1986	3262	7 901	11 557	1239	3 961	665	2330	5318	2846	39 079
1987	3578	7 329	2 941	824	3 294	792	1300	3884	785	24 727
1988	3625	4 182	19 348	1046	2 259	404	350	4796	866	36 876
1989	3543	7 103	713	195	10 012	440	5647	5395	2773	35 821
1990	5920	4 511	7 503	703	7 587	584	3563	4566	3405	38 342
1991	4082	16 483	1 513	814	6 504	822	2728	6488	1111	40 545
1992	2311	7 234	3 885	1610	4 153	865	1014	4033	334	25 439
1993	3673	7 002	714	425	12 695	1985	584	3837	3140	34 055

March 29, 1996, DFO in the so-called Mifflin Plan incorporated some of these options in a major restructuring of the industry. The plan involves an $80 million buy-back of salmon licences, a licensing requirement that vessels use only one type of gear, a system of area licensing, and a provision for the stacking of salmon licences (DFO 1996).

We propose another option that has the some of the features of both area licensing and ITQs but is a different form of rights-based management. Our proposal does not address the management of aboriginal fisheries and co-management, which is examined in detail by Pinkerton (1994) and involves the assignment of bidding rights called fishers' individual salmon harvesting rights (FISHRs) to individual fishers. These rights would be annually exhaustible, assigned gratis, and used by fishers to bid for the right to participate at salmon openings where the number of vessels permitted to fish would be fixed in advance by the regulator. The setting of the total number of vessels, but not which vessels, that participate at salmon openings is a major benefit of FISHRs, as it would provide a much greater degree of control over the resource. This is crucial in the salmon fishery where the total number of salmon can fluctuate a great deal from year to year and from area to area (Copes (1986) pointed out the difficulty of applying ITQs in a rapidly fluctuating fishery). Thus, they would help DFO to focus on overall escapement and indirectly fishing effort while allowing fishers to decide where to fish.

The British Columbia salmon fishery

Salmon are caught by aboriginal, sport, and commerical fishers. The commercial fishery is restricted to three gear types: troll fishing using poles and lines, gill nets that trap fish, and seine nets that are laid by vessels that encircle schools of salmon. At the start of 1996, there were 4439 licensed salmon vessels of which only 545 were allowed to use seine gear while the rest were able to use troll or gill-net gear. Many of the commercial vessels are also licensed in other fisheries. In general, the troll vessels exert less fishing power per vessel than seiners and are allowed to fish for longer periods. Troll vessels are also the first to harvest the returning runs followed by the seiners and gill netters. All vessel types are restricted to fishing in defined salmon openings that vary from hours to days depending on the season, location, and gear type used to catch the fish.

An important feature of the salmon fishery is not only the return of fish to spawn in the rivers but the fluctuations in the numbers of returning salmon. For example, in 1987 the number of sockeye caught per days fished was 743 565 for troll vessels and in 1988 it was 244 352, a threefold decrease in just 1 year (DFO 1988, 1989). Even greater variations exist for individual openings by area and time of year (Table 1). For instance, the catch of salmon in DFO regions 6–8 ranged from 19.3 million fish in 1988 to just 713 000 in 1989, a 27-fold decrease (a map of the fishing regions is given in DFO 1993, pp. 2–3). There is also considerable variation in the species mix. The proportion of sockeye salmon of the total catch in DFO area 1–2W fluctuated from 2.5% of the total in 1985 to 35% in 1993.

Wide variations in overall salmon catches by area and fluctuations over time (Fig. 1) and the catch levels by area relative to the average harvest over all areas over the same period (Fig. 2) illustrate the difficulties of managing the salmon fisheries and the uncertainty faced by fishers. The history of the industry shows that in good years, high returns of salmon tend to encourage investment (The DPA Group Inc. 1988) in vessels and gear and, in the poor years, there is little opportunity to redirect fishing effort elsewhere. Thus, over time the commercial fleet capacity to harvest the resource has increased, making fishers more vulnerable to changes in market conditions and decreases in salmon runs.

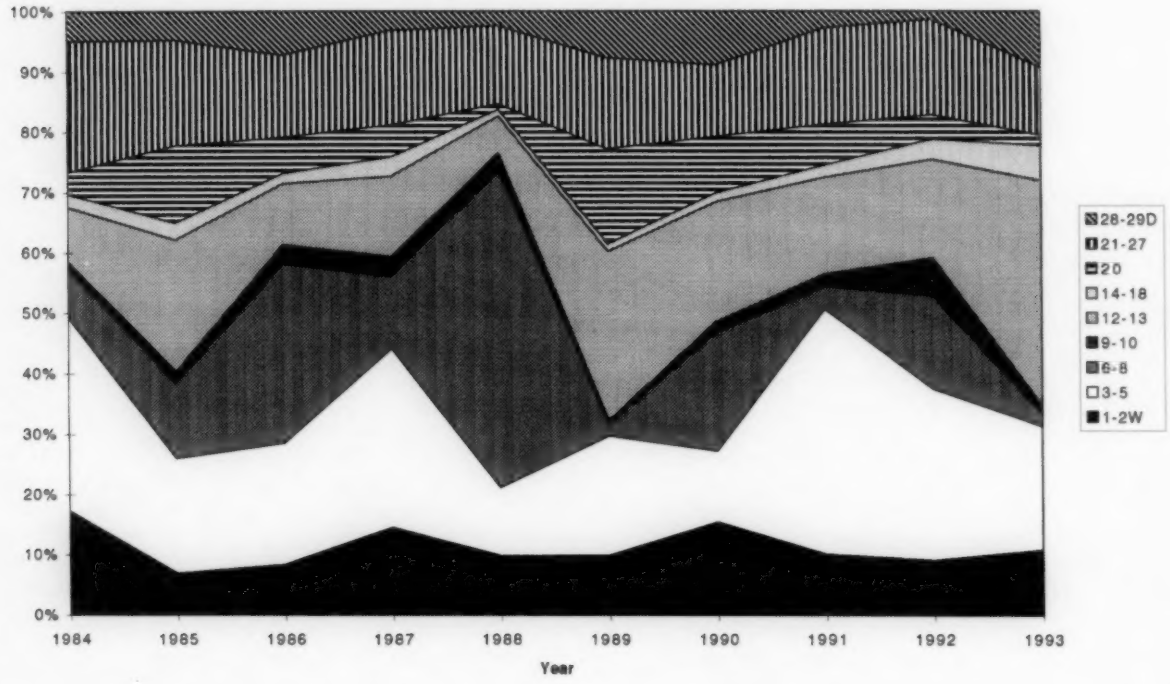
History of regulation

The most cost-effective method to harvest salmon is to wait until they migrate up the river system and use traps, weirs, or nets to catch them. Because it is possible to block an entire river mouth, it is possible to harvest an entire run. In the early years of fisheries regulation, this led to the eventual prohibition on certain traps and weirs that were traditional methods of harvesting by aboriginals. In time, restrictions have been placed on many aspects of fishing, including season length, vessel size, permitted gear type, and the number of vessels allowed to harvest the resource.

As early as 1890, net and gear restrictions, vessel licensing, and closures were established to protect Fraser River salmon runs. This focus on controlling input and effort of fishers still continues. Limited entry, which restricts the total number of vessels permitted to fish, was introduced throughout British

© 1997 NRC Canada

Fig. 1. Percentage of overall salmon catch from each area, 1984–1993 (DFO 1993).



Columbia in 1969. Along with limits on vessel numbers, a program of buying back salmon licences was implemented from 1971 to 1974. This retired 362 vessels from the salmon fleet at a cost of $6 million. A much smaller buy-back program was also introduced in 1981 that retired 26 vessels at a cost of $2.5 million (Pearse 1982).

Despite restrictions on the number of vessels, competition among fishers for the limited catch has led to greater fishing power per vessel (Wilen 1989). To address this problem, the regulator has imposed other restrictions, including a prohibition on stacking of licences, which was used by fishers to increase the licensed tonnage on particular vessels (Pearse 1982; Schwindt 1987; Fraser 1979). These regulations and restrictions on net sizes and gear type have, unfortunately, not prevented fishers from increasing the fishing power of their vessels and dissipating the returns from the fishery (for a general review of licensing in fisheries, see Townsend 1990). For example, seine vessels, which in the past were only capable of making four sets of their net per day, are today able to make over 25 sets per day (DFO 1995*b*). In response, the regulator has reduced the number of fishing days so as to ensure sufficient escapement and the sustainability of the fisheries. For instance, in area 12 in Johnstone Strait between Vancouver Island and the mainland of British Columbia, seine vessels in 1972 could fish for salmon for 51 days, while in 1994 they were permitted to fish for only 3.25 days (DFO 1995*b*). Unfortunately, reduced opening days increases the pressure on fishers to increase their fishing power so as to be able to catch their expected harvest in the limited time available to them (Clark 1982). Consequently, fisheries management has become increasingly difficult in that a mistake in estimating the escapement or setting the length of an opening could devastate the viability of future salmon runs.

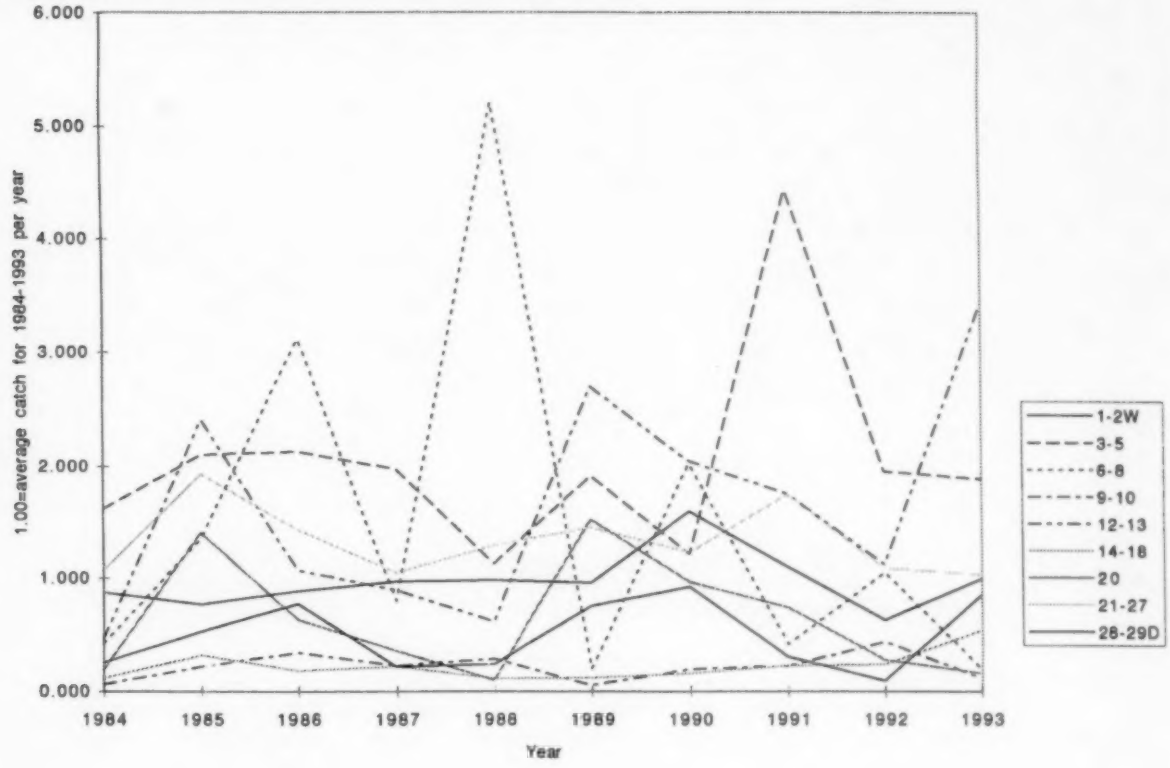
The economic consequences of excess competition and "racing behavior" to catch the limited harvest of salmon by fishers are considerable (Scott 1986). Dupont (1990) estimated that past management has dissipated most of the potential rent. Dupont (1990) found that removing the redundant vessels from the salmon fleet would have increased the rent by some $52 million in 1982 dollars, although rents would increase by another $20 million if the minimum number of vessels harvesting salmon was at its optimal size. There are also increased costs that arise from substitution of unregulated for regulated inputs (Munro and Scott 1985), excess mobility and movement (Wilen 1989), and congestion that can arise when many vessels attempt to harvest salmon at openings that are limited in terms of area and time. It is against this background of increasing fishing power over time, declines in some salmon runs, and low incomes of fishers that various management options must be considered.

Management options for the salmon fisheries

The current management system is based on effort and input controls and encompasses a complex set of regulations. These controls have, until recently, been effective at ensuring the continued sustainability of the fishery but have been ineffective at controlling excess investment and declining returns in the fishery. Without changes in current management, the regulator may be unable to ensure sufficient escapement in the face of uncertainties over the size of

© 1997 NRC Canada

Fig. 2. Relative catch levels for the different areas, 1984–1993 (DFO 1993).



salmon runs and will be unable to improve the financial viability of the fleet.

A recognition of the need to change existing fisheries management has led to a review of various alternatives by the regulator in collaboration with stakeholders in the industry. The objectives of the management changes are to ensure the sustainability of the resource, a reasonable standard of living for fishers, and increased participation by stakeholders in regulating the salmon fisheries (DFO 1995*a*). Possible management options include (*i*) increased effort controls, (*ii*) area licensing, (*iii*) single-gear licensing, (*iv*) buy-back of licences, (*v*) separate licence management, (*vi*) double licensing, and (*vii*) ITQs. The restructuring plan announced by DFO on March 29, 1996, the so-called Mifflin Plan, involves an $80 million licence buy-back, single-gear controls that restrict gill-net and troll vessels to use only one gear type in each area, a provision for the stacking of fish licences, and a system of area licensing that restricts seine vessels to one of two areas and gillnet and troll vessels to one of three but different locations. A feature of the area licensing under the Mifflin Plan is that fishers can choose the area where their licence will permit them to fish but it will be fixed for 4 years.

Each of the seven potential management options is briefly described below and evaluated in terms of the following criteria: (*i*) sustainability — the ability to improve control over harvests at individual openings; (*ii*) equity — the perception of equal treatment and fairness among fishers; (*iii*) short-run rent dissipation — the ability to reduce the excess use and misallocation of variable inputs that arises from racing behavior, excess mobility as fishers move from opening to opening, and congestion costs that arise when vessels interfere with the fishing operations of others; and (*iv*) long-run rent dissipation — the ability to limit future excess capital investment by fishers and reduce the number of vessels in the fleet.

Increased effort controls

Increased effort controls on gear and vessels represent a continuation of past policies. Unfortunately, salmon fishers have been particularly good at using nonregulated inputs or adjusting their behavior so as to continue to increase fishing effort to catch a greater share of the catch. On the grounds of equity, this option is potentially neutral if applied to all fishers and gear types uniformly. The history of the Pacific salmon industry, however, suggests that it would fail to address the problems of short-run or long-run rent dissipation. Effort controls, short of restricting the number of vessels per opening, would fail to help the regulator to control escapement or to determine the length of openings or increase the safety margin faced by regulators in ensuring sustainability of salmon runs.

Area licensing

Area licensing has been applied with some success in the herring roe fisheries in British Columbia (MacGillivray 1986)

© 1997 NRC Canada

and would place restrictions on where fishers may harvest salmon. Given the large fluctuations in harvests of salmon by area, such a system is unlikely to be equitable in that it may restrict some fishers to poor harvests while other, more fortunate fishers enjoy better salmon runs. Transferability of area licences would, however, mitigate the problem as would allowing ownership of multiple licences (Wilen 1989). Depending on the size of the areas defined by the regulator and the number of vessels assigned, area licensing should reduce the number of fishers at openings. In turn, this should increase the margin of safety for DFO when setting length of openings and may reduce congestion costs. In the Mifflin Plan, however, there are only two areas for seiners and three areas for troll and gill-net vessels, which suggests that the problem of congestion and control will not be resolved. Only if the number of vessels at each opening is small enough with area licensing and if fishers perceive the licence as a durable property right is there likely to be a reduction in the race to fish and excess mobility and increased cooperation among fishers (Yamamoto 1995; Wade 1987). In turn, this may reduce both short- and long-run rent dissipation.

Single-gear licensing

This option would restrict all licence holders to just one gear type and seeks to reduce the fishing effort applied by the salmon fleet. This policy is not equitable in that some fishers use a mix of gill nets and troll to catch salmon. As with any conventional system of vessel licensing, this will fail to limit excessive levels of fishing effort and prevent rent dissipation (Clark 1980). Single-gear licensing would also fail to reduce congestion costs at openings or help regulators manage openings to ensure sufficient escapement. Under the Mifflin Plan, troll and gill-net vessels will be restricted to the use of just one gear in each area of the coast if they only own one licence.

Buy-back of licences

A succesful buy-back of licences would reduce the number of vessels and thus the fleet redundancy in salmon harvesting. It has the potential, therefore, of increasing the returns of those fishers who remain active in the fishery. A reduction in the number of vessels would also facilitate management of openings for escapement. It is, however, only an indirect control on the fishing effort applied at each opening. If open to all fishers of any gear type, a buy-back of licences is equitable to fishers if paid for by the regulator. If the buy-back is financed by the fishers who remain, then it favors the fishers who sell their licences and exit the fishery. The principal failing of a buy-back is that by itself, it fails to change the incentives faced by fishers and tends to remove the less effective fishers and vessels from the fishery. Over time, those fishers who do not accept the buy-out may substitute nonregulated for regulated inputs and there may be excess mobility as vessels move from one opening to the next in a rush to fish.

Separate licence management

This option changes the current division of salmon licences based on seine or gill-net and troll vessels by placing the seine licences in two categories. Vessels in one category would be prevented from fishing at openings reserved for the other. Separate licensing is inequitable, as it penalizes seiners compared with trollers and gill netters. It also fails to change the incentives of fishers and does nothing to prevent short- or long-run rent dissipation. In fact, by restricting seiners to a smaller number of openings, there may be increased incentives to invest in extra inputs so as to maintain current catches at the expense of other fishers. Reducing the number of seiners at particular openings may, at least in the short run, increase the safety margin of DFO in setting opening times and escapement levels.

Double licensing

Double licensing would require that vessels have two fishing licences on one vessel to be able to participate in the fishery. It would significantly reduce the total number of vessels in the fleet and would represent an industry-funded buy-back program of licences. Double licensing is, however, inequitable, as some fishers currently do own multiple licences and licence values are different for seiners compared with troll and gill-net licences. Although it has the potential to significantly decrease the total number of vessels in the fleet, it would most certainly not reduce the total fishing effort by the same proportion. This is because the vessels that would exit the fishery would almost certainly be the older and less competitive boats. As with the buy-back option, double licensing fails to change the fundamental incentive for fishers to compete among themselves, which leads to excess fishing effort.

Individual transferable quotas

ITQs would allocate shares of the harvest to individual vessels. Transferability of a right to harvest a share of the salmon catch could result in a considerable rationalization of the fleet and, in time, a reduction in the size of the fleet. This approach, by providing fishers with a greater assurance of catching a given quantity of salmon, provides greater incentive for fishers to minimize costs. Thus, ITQs have the potential to reduce short- and long-run rent dissipation in the salmon industry as they have done in other fisheries (Grafton 1996; Squires et al. 1995). ITQs would not, however, provide a complete property right because the salmon fisheries would still be owned by the people of Canada (Scott 1985).

A major issue with ITQs is how to apply them in salmon fisheries where estimates of the total allowable catch vary over the fishing season and harvests are highly variable. To implement ITQs, fishers would be given rights either to a portion of the total harvest or to a share of individual salmon runs after sufficient escapement. The problem is that the allowable harvest is not known until the end of the season and estimates of what should be caught change over the season. In addition, monitoring the catch of over 4000 vessels fishing throughout British Columbia over just a few months or even weeks would be extremely difficult and costly. Further, the principal advantage that arises from ITQs is that they provide a much greater assurance to fishers that they will harvest a given share of the catch. This assurance and the fact that quotas are scarce provides incentives for fishers to minimize their costs for a given harvest level. In the salmon fishery, however, where there is considerable uncertainty over escapement, fishers may be tempted to catch as much as they could at the beginning of the season. This could lead to further congestion costs and would reduce the potential gains.

© 1997 NRC Canada

Fishers' individual salmon harvesting rights

An alternative management option for the British Columbia salmon industry are FISHRs, which are transferable property rights in the form of *bidding* rights for fishers. These rights would be annually exhaustible but renewable and would allow fishers to bid for the right to participate at salmon openings where the number of vessels is determined in advance by the regulator. FISHRs share a feature with ITQs in that they are property rights that are transferable and thus have the potential to rationalize the salmon fleet. They share a feature with area licensing in that, by themselves, they do not restrict the catch of vessels but rather the number of vessels at salmon openings. This is important in an overcapitalized fishery where a small error in the length of a salmon opening and escapement could have catastrophic consequences for the fishery. By allocating bidding rights to individuals and companies, however, FISHRs do not "micromanage" the fishery, because it is the participants themselves who would determine which vessels fish at which opening.

Using the characteristics of property rights defined by Scott (1986), we review the features of FISHRs. They represent an *exclusive* right insofar that no one can participate at a salmon opening without successfully bidding for the right. FISHRs would also be fully *transferable* and *divisible* among salmon licence holders. For example, it would be possible for an owner of a vessel with 20 FISHRs to sell 1 FISHR to 20 separate salmon licence holders. These rights would be annually exhaustible but would be *durable* in the sense that the bidding rights would be renewed every year. The *quality of title* to FISHRs would be attenuated to the extent that they represent a right to bid and not a right to a fixed number of salmon and because they would be attached to salmon licences that exist at the discretion of the Minister of Fisheries and Oceans. FISHRs would also be a *flexible* property right in the sense they could accomodate significant changes in both the size of the salmon stocks and changes in fisheries regulations in terms of licensing and input controls, as well as circumstances of individual owners.

FISHRs have the potential to allow the regulator to reduce the number of vessels, increase the return to fishers, and improve the regulator's control over fishing effort. FISHRs may also help in the allocation of fish across competing users. Thus, they may be able to meet the stated management objectives for the salmon fisheries — sustainability, industry viability, and partnership between the regulator and users — and have the potential to be equitable and reduce short- and long-run rent dissipation.

In comparison with other management options, such as government-funded licence buy-backs, FISHRs would not involve a major expenditure for the regulator. They could also simplify fisheries management relative to the option of increasing effort controls, which would require further restrictions on fisher behavior. FISHRs would also provide much greater flexibility to fishers than would area licensing and would be easier to monitor and control than ITQs, which require verification of the catch and landings of vessels.

Modus operandi

A system of FISHRs could operate in the following way. First, the regulator would determine the desirable number of vessels that would be allowed at a particular salmon opening. This number would be set at a level to ensure that the regulator has adequate control and a safety margin to ensure adequate escapement. Determining the desirable number of vessels per opening would not be easy and could be influenced by lobbying from fishers unsuccessful in the bidding process. It would, however, be an improvment over current practice.

The number of permitted vessels would be announced along with a reserve price in terms of FISHRs required to be a successful bidder. Second, all salmon licence holders would have the opportunity to bid the number of FISHRs they would be prepared to pay to participate in the specified opening. This bidding process should be as flexible as possible. The regulator could, for example, have a telephone number with multiple lines that would allow fishers to dial and specify the number of FISHRs they would be prepared to pay. The service could also supply up-to-date information on openings, the deadline for bids, the reserve price, the number of vessels to be allowed, and the successful bidders. Thus, fishers could bid at sea and at any time. To ensure that fishers understand the bidding process, it would probably be best to sequentially auction over the season the right to participate at salmon openings. This would also permit fishers to learn from past bids and help them to prioritize their bidding. Such a system would also be much easier to implement than more complicated auctions such as the sequential ascending auction used in the sale of broadcast spectrum licences in the United States (McAfee and McMillan 1996). The bidding for FISHRs would also have the unusual characteristic that all successful bidders, irrespective of the number of FISHRs paid, would be treated identically in terms of access to the salmon. More commonly, as in the U.S. offshore oil and gas lease auctions, the winner takes all (Porter 1995). An advantage of using FISHRs for bidding rather than money is that they provide a capital gain to initial holders of the rights, which should help in their acceptance by fishers. Further, as capital markets are imperfect, up-front cash payments to successfully bid for the right to participate in salmon openings could be particularly difficult for independent owner–operators with smaller vessels.

Once the deadline for the bids at a particular opening is past, then if there were x number of vessels deemed desirable at the opening, the top x number of bids would secure the right to fish. At the opening, it would be easy to determine the commercial fishing vessels with the right to fish in that all successful bidders would be required to submit their licence and commercial fishing vessel number. FISHRs would be annually exhaustible such that rights would be renewed every year but unused rights from previous years would not be permitted to be carried over.

Although the regulator would determine overall catch or escapement levels, as well as the number of vessels able to participate, it would be the fishers themselves through their bidding who would determine where they would fish. For the system to work, the number of vessels permitted at a particular opening would have to be less than the number of vessels willing to participate. Given the excess harvesting capacity that currently exists (Pearse 1982), such a condition is likely to be easily met. Under this constraint, fishers would try to concentrate their bidding at those openings that would suit their preference or comparative advantage. This, coupled with the fact

© 1997 NRC Canada

that the number of vessels participating at an opening would be known, could reduce racing behavior among fishers. If an opening were such that the specified number of vessels were small enough, then fishers could even act cooperatively.

Allocation

A major question in establishing any set of property rights in fisheries is the initial allocation. Whatever the method chosen, the rights should be allocated to ensure some industry support for FISHRs. In determining the allocation, the regulator may also wish to consider historical participation in the fishery, gear type, vessel size, and any other factors deemed appropriate. Given past conflicts in the Pacific salmon fisheries, whichever method of allocation is chosen will probably lead to complaints from some fishers or groups. One method that would minimize conflicts would be to assign FISHRs, equally to all salmon licence holders. The regulator could also split the total number of FISHRs among the three gear types based on their catch of the total salmon harvest. For example, trollers, which account for about 30% of the total catch of salmon, would receive in total 30% of the FISHRs which would then be distributed equally among all troll vessels, while seiners would receive an equal allocation of FISHRs based on their 40% share of the total catch. Because FISHRs are bidding rights, however, an equal allocation across vessels would not result in an equal catch, as some seiners exert much greater fishing power than do gill-net or troll vessels.

Transferability

It would be important to allow transferability of FISHRs among salmon licence holders. It is transferability that would permit a rationalization of the industry over time. This rationalization would arise whenever fishers *voluntarily* sold their licence and FISHRs to other salmon licence holders and, unlike a government-sponsored buy-back of vessels, would be paid for entirely by fishers themselves. Thus, the most profitable fishers should be able to purchase rights from other fishers and increase their share of the total harvest of salmon without necessarily increasing their fishing effort. Transferability of FISHRs would be important to allow the structure of the industry to adapt to changing market conditions and changes in efficiency across gear types. Thus, transferability of FISHRs across gear types should be considered, especially in schooling fisheries such as salmon, where there may be significant economies of scale (Wilen 1989; Dupont 1990). To prevent excessive market power (Anderson 1991) in trades of FISHRs, limits would need to be placed on the total share owned or leased by a company or individual. Trades across gear types may also have negative impacts on some fishing communites, which may also need to be considered by the regulator.

Regulator's role

The regulator would have two instruments at its disposal. The first would be a determination of the number of vessels permitted to participate at openings, and the second would be a reserve bid for the right to participate at an opening. The purpose of the reserve bid would be to counter strategic behavior. Although such problems could never be completely resolved, minimum bids would at least place constraints on the ability of firms to engage in rigging bids. The regulator may also wish to keep some input controls such as limits on net sizes to ensure the sustainability of the resource.

Another objective the regulator may wish to implement is a more or less constant "vessel to expected harvest" ratio at openings. Theoretically, this would mean that a priori fishers would know that they would have an equal expected chance of success in any opening. Thus, fishers could then choose to participate in the openings where they perceive a comparative advantage without having to gauge possible changes in the behavior of the regulator or other fishers over the course of the season.

Advantages and disadvantages

Fishers' individual salmon harvesting offers a number of advantages over some of the options in the Mifflin Plan, such as area licensing. First, fishers would be able to operate in any area and opening they wish, provided they were willing to pay the price in FISHRs. If conditions and the size of runs changed in certain areas over time, then fishers would be able adjust their behavior much more easily than with area licences. This is important in the salmon fishery, which is subject to substantial fluctuations in the harvest. Second, the regulator would not only be able to determine the number of vessels allowed to fish in a given area but also specific openings within areas. This would provide added flexibility to ensure the sustainability of stocks. It would not, however, require that the regulator determine which vessels would participate in a particular opening. Because FISHRs are transferable and could be used to bid on any opening, they would also provide an equitable means for the regulator to change harvest levels over time or throughout an area without having to reallocate rights on an annual basis.

Another desirable feature of FISHRs is that they let fishers decide where and how they should fish through bidding for the right to participate at salmon openings. In this sense, it is a step away from "micromanagement" but still ensuring that the regulator maintains control over the stocks by setting escapment levels and indirectly through determining the number of vessels at openings. Over time, this should increase net returns in the fishery in that tradeability of FISHRs should allow more profitable fishers to buy out those with lower returns. If this is combined with some rental charge on FISHRs, the regulator should be able to generate returns from the fishery for the benefit of the owners, the people of Canada. If not, the expected rent from the fishery would go to the first generation of holders of FISHRs. One potential method of rent capture would be to base the charge as a percentage of the current market price of FISHRs, although there are several alternatives such as a royalty charge or licence fees (Grafton 1995).

As with any fishery management scheme, there are also some drawbacks with this system. First, it requires detailed in-season management by the regulator in terms of setting the number of vessels at each opening. Second, FISHRs require that individuals be actively involved in bidding and bear the risk of spending their rights on an opening that may provide less fish than expected. In the past, the equal right to participate in all openings became the de facto means for handling that uncertainty. Some fishers would, no doubt, take some time to adjust to the new management and may be unhappy with the risks of bidding on an uncertain outcome. Fishers may also be concerned about having unused bidding rights at

© 1997 NRC Canada

the end of the season or even missing out entirely from participating at openings. This may lead to higher bids at the start of the season. However, if the regulator has determined to reduce the number of vessels that can participate in any opening, it appears that fishers will face more risk through restricted opportunities regardless of the management scheme.

This system extends fishers more decision-making power, which may result in improvements in efficiency in return for increased risk through the bidding process. By restricting the number of vessels per opening and allowing transferability of FISHRs, the level of fishing effort, excess mobility, and racing behavior should be reduced. If the number of vessels permitted at openings, set by the regulator, was also significantly reduced, it would also decrease congestion costs. Smaller numbers at openings may also lead to cooperation among fishers, which can help reduce racing behavior among fishers. Unlike individual quotas, however, FISHRs without accompanying catch limits would not restrict the amount of fish that vessels could harvest at a particular opening. Thus, the incentive may still exist for fishers to compete among themselves and perhaps invest in vessels and equipment that may be privately beneficial but which would not increase the total return from the fishery.

A variant of FISHR management that could help address the incentive of fishers to race to fish would be to specify not only the number of vessels at openings but vessel catch limits based on the manager's desired escapement and the bids received. For example, a fisher who successfully bid a number of FISHRs equal to 5% of the total paid by all successful bidders would be allocated a catch limit of 5% of the maximim desired catch at the opening. To ensure flexibility, the allocated catches to fishers could be transferable but only among the successful bidders at the specified opening and only after notifying the regulator. Specifying catch limits to succesful bidders prorated on their proportion of the total number of FISHRs paid would, however, be more difficult to enforce and manage than merely determining which vessels have the right to participate in an opening. Thus, using FISHRs alone or FISHRs combined with output controls represents a trade-off between simplicity and lower monitoring and enforcement costs and a greater control over the activities of vessels.

Combining output controls with bidding rights to participate at salmon openings illustrates another advantage with FISHRs: their flexibility. For example, it would be possible to bundle openings into portfolios so that fishers could bid on several at once. FISHRs also are highly divisible, which would permit both small and large operators to benefit from trading.

Concluding remarks

The salmon fisheries of British Columbia face a number of crises: jurisdictional and catch disputes with the United States, a decline in some salmon species such as sockeye and chinook, conflicts between aboriginal, commercial, and sport fishers, and continuing excess capacity. Improved management will not address all of these problems but has the potential to improve the sustainability and viability of the industry. FISHRs, which allow fishers to bid for the right to participate at salmon openings where the number of vessels is fixed by the regulator, are one management option worthy of consideration. They offer the potential to equitably rationalize the salmon fleet, increase the share of the harvest by more profitable vessels, reduce congestion costs, and increase cooperation among fishers.

References

Anderson, L.G. 1991. A note on market power in ITQ fisheries. J. Environ. Econ. Manage. **21**: 292–296.

Christy, F.T., Jr. 1973. Fishermen's quotas: a tentative suggestion for domestic management. Univ. R.I. Law Sea Inst. Occas. Pap. No. 19.

Clark, C.W. 1980. Towards a predictive model for the economic regulation of commercial fisheries. Can. J. Fish. Aquat. Sci. **37**: 1111–1129.

Clark, C.W. 1982. Models of fishery regulation. *In* Essays in the economics of renewable resources. *Edited by* L.J. Mirman and D.F. Spulber. North-Holland, Amsterdam, The Netherlands. pp. 273–287.

Copes, P. 1986. A critical review of the individual quota as a device in fisheries management. Land Econ. **62**: 278–291.

DFO. 1988. British Columbia commercial catch statistics by species, gear, month and area. Department of Fisheries and Oceans, Vancouver, B.C.

DFO. 1989. British Columbia commercial catch statistics by species, gear, month and area. Department of Fisheries and Oceans, Vancouver, B.C.

DFO. 1993. Annual summary of British Columbia catch statistics 1993. Department of Fisheries and Oceans, Vancouver, B.C.

DFO. 1995*a*. Options for commercial fisheries management. April 1995. Department of Fisheries and Oceans, Vancouver, B.C.

DFO. 1995*b*. Summary discussion paper. Prepared for the Pacific Policy Roundtable. September 1995. Department of Fisheries and Oceans, Vancouver, B.C.

DFO. 1996. Minister announces plan to revitalize salmon fishery. March 29. NR-PR-96-15E. Department of Fisheries and Oceans, Vancouver, B.C.

Dupont, D. 1990. Rent dissipation in restricted access fisheries. J. Environ. Econ. Manage. **19**: 26–44.

Fraser, G.A. 1979. Limited entry experience of the British Columbia salmon fishery. J. Fish. Res. Board Can. **36**: 754–763.

Fraser River Sockeye Public Review Board. 1995. Fraser River sockeye 1994: problems and discrepancies. Catalogue No. Fs23-263/1995E, Public Works and Government Services Canada, Ottawa, Ont.

Grafton, R.Q. 1995. Rent capture in a rights based fishery. J. Environ. Econ. Manage. **28**: 48–67.

Grafton, R.Q. 1996. Individual transferable quotas: theory and practice. Rev. Fish Biol. Fish. **6**: 5–20.

Hatfield Consultants Ltd. 1996. The Strait of Georgia fisheries sustainability review. Vancouver, B.C.

Healey, M.C. 1993. The management of pacific salmon fisheries in British Columbia. *In* Perspectives on Canadian marine fisheries management. *Edited by* L.S. Parsons and W.H. Lear. Can. Bull. Fish. Aquat. Sci. **226**: 243–266.

MacGillivray, P. 1986. Evaluation of area licensing in the British Columbia roe herring fishery 1981–1985. *In* Fisheries control programs worldwide. *Edited by* N. Mollett. University of Alaska, Fairbanks, Alaska. pp. 251–274.

McAfee, R.P., and McMillan, J. 1996. Analyzing the airwaves auction. J. Econ. Perspect. **10**: 159–175.

Moloney, D.G., and Pearse, P.H. 1979. Quantitative rights as an instrument for regulating commercial fisheries. J. Fish. Res. Board Can. **36**: 859–866.

Munro, G.R., and Scott, A.D. 1985. The economics of fisheries management. *In* Handbook of natural resource and energy economics. Vol. II. *Edited by* A.V. Kneese and J.L. Sweeny. North-Holland, Amsterdam, The Netherlands. pp. 623–676.

© 1997 NRC Canada

Pearse, P.H. 1982. Turning the tide: a new policy for Canada's Pacific fisheries. The Commission on Pacific Fisheries Policy Final Report. Catalogue No. Fs23-18/1982E, Supply and Services Canada, Ottawa, Ont.

Pinkerton, E.H. 1994. Local fisheries co-management: a review of international experiences and their implications for salmon management in British Columbia. Can. J. Fish. Aquat. Sci. **51**: 2363–2378.

Porter, R.H. 1995. The role of information in U.S. offshore oil and gas lease auctions. Econometrica, **63**: 1–27.

Schwindt, R. 1987. Public policy and Pacific salmon's harvesting crisis. *In* Resource rents and public policy in western Canada. *Edited by* T. Gunton and J. Richards. The Institute for Research on Public Policy, Ottawa, Ont. pp. 215–248.

Scott, A.T. 1985. Property rights: developing characteristics of interests in natural resources. *In* Progress in natural resource economics. *Edited by* A.T. Scott. Clarendon Press, Cambridge. pp. 377–401.

Scott, A.T. 1986. Catch quotas and shares in the fishstock as property rights. *In* Natural resource economics and policy applications. *Edited by* E. Miles, R. Pealy, and R. Stokes. University of Washington Press, Seattle, Wash. pp. 61–96.

Squires, D., Kirkley, J., and Tisdell, C. 1995. Individual transferable quotas as a fisheries management tool. Rev. Fish. Sci. **3**: 141–169.

The DPA Group Inc. 1988. British Columbia salmon fleet financial performance 1981–1985. Report prepared for the Department of Fisheries and Oceans, Pacific Region, Vancouver, B.C.

Townsend, R.E. 1990. Entry restrictions in the fishery: a survey of the evidence. Land Econ. **66**: 359–378.

Wade, R. 1987. The management of common property resources: collective action as an alternative to privatisation or state regulation. Camb. J. Econ. **11**: 95–106.

Wilen, J.E. 1989. Rent generation in limited entry fisheries. *In* Rights based fishing. *Edited by* P.A. Neher, R. Arnason, and N. Mollett. Kluwer Academic Publishers, Dordrecht, The Netherlands. pp. 249–262.

Yamamoto, T. 1995. Development of a community-based fishery management system in Japan. Mar. Res. Econ. **10**: 21–34.

© 1997 NRC Canada

PERSPECTIVE

Are causes knowable? Some consequences of successional versus toxicological interpretations of the Great Lakes Water Quality Agreement[1]

Michael Gilbertson

Abstract: The focus of the present Great Lakes Water Quality Agreement between Canada and the United States is on transboundary pollution and particularly the injury to health and property from exposures to persistent toxic substances. Several scientists and policy advisors have suggested, on the basis of the Ecosystem Approach and systems theory, that the agenda should move on to other demanding topics, such as the introduction of exotic species, biodiversity, habitat restoration, fisheries management, sustainable development, and governance of the Great Lakes basin. None of these issues, however, poses the same scale of threat to national sovereignty and bilateral interests as does transboundary pollution. Thus, none warrants the degree of attention that has been and should be paid, despite declining budgets, to assessing and controlling releases of persistent toxic substances that continue to injure fish, wildlife, and human health. Instead, it is recommended that other existing institutions and treaties, such as the several commissions in the Great Lakes basin, as well as local, state, and provincial governments, should address these other pressing issues through the development of other bilateral or multilateral agreements and improved institutional arrangements.

Résumé : L'Accord relatif à la qualité de l'eau des Grands Lacs entre le Canada et les États-Unis est actuellement centré sur la pollution transfrontalière et, en particulier, les dommages à la santé et à la propriété causés par l'exposition à des substances toxiques persistantes. Plusieurs scientifiques et plusieurs conseillers en matière de politiques ont proposé, en se basant sur l'approche écosystémique et la théorie des systèmes, que le programme fasse place à d'autres sujets bien en vue comme l'introduction d'espèces exotiques, la biodiversité, la restauration des habitats, la gestion des pêches, le développement durable et la gestion du bassin des Grands Lacs. Toutefois, aucune de ces questions ne menace autant la souveraineté nationale et les intérêts des deux parties que la pollution transfrontalière. Par conséquent, aucune d'entre elles ne mérite le niveau d'attention porté, à juste titre et malgré les réductions budgétaires, au contrôle et à la limitation des rejets de substances toxiques persistantes qui continuent de nuire à la santé des poissons, de la faune et des humains. Nous recommandons plutôt que d'autres institutions et traités, comme les nombreuses commissions du bassin des Grands Lacs ainsi que les gouvernements locaux, étatiques et provinciaux s'occupent de ces autres questions par l'élaboration d'autres accords bilatéraux ou multilatéraux et de mesures institutionnelles améliorées.

International progress on Great Lakes water quality

Canada and the United States have made significant progress, during the past 30 years, in improving water quality in the Great Lakes basin. Much of the success is attributable to the growth of bilateral cooperation on research and regulatory matters that occurred in the 1960s and 1970s. This cooperation was initiated through the 1964 reference, under the 1909 Boundary Waters Treaty, to the International Joint Commission concerning the pollution of the lower Great Lakes (International Joint Commission 1969). Before the reference, there had been little bilateral communication despite the severity of the pollution in the boundary waters, particularly the interconnecting channels such as the Detroit River and the Niagara River. The completion of the reference in 1969 led directly to the 1972 signing of the Great Lakes Water Quality Agreement that responded to the decades of deteriorating water quality resulting from pollution by municipal, agricultural, and industrial wastes. The focus was not only on the control of a variety

Received March 15, 1996. Accepted November 18, 1996.
J13340

M. Gilbertson. International Joint Commission, 100 Ouellette Avenue, Windsor, ON N9A 6T3, Canada. e-mail: gilbertsonm@ijc.wincom.net

[1] The views expressed in this article are those of the author and should not be construed to be necessarily those of the International Joint Commission.

© 1997 NRC Canada

of pollutants that had been well characterized, such as nitrates, ammonia, oil and grease, phenols, and suspended solids that caused effects at the local level, but also on negotiated reductions in the loadings of phosphorus to each of the Great Lakes, since it was believed that eutrophication could be controlled by limiting phosphorus concentrations in the lakes. The development of water quality objectives derived from aquatic toxicology was an important strategy in the bilateral efforts to provide a scientifically defensible rationale to reduce pollution. This traditional water quality approach remains a cornerstone of efforts to maintain and restore the integrity of the waters of the Great Lakes. It is primarily practiced by analytical chemists, aquatic toxicologists, and engineers. Measures of compliance are in terms of comparisons of the results of chemical or microbiological analyses of water or effluent samples with prescribed standards. Biological indicators of compliance generally include an assessment of assemblages of invertebrates.

The Agreement was signed during a period of rising environmental concern, and conferred mutual benefits on both parties. The Agreement contained a reference to the International Joint Commission to assist the parties in implementing the provisions of the Agreement and terms of reference to set up a Great Lakes Water Quality Board composed mainly of regulatory officials and a Great Lakes Research Advisory Board composed of government research managers, as well as a directive to establish a Great Lakes Regional Office to undertake secretariat functions. These new organizations brought immediate improvements in the consultative process and transfer of information among the two parties and their jurisdictions. In the United States, there was a growing public consensus that water quality in the nation had become unacceptable and that a strong regulatory initiative was required at the national level. From the Great Lakes states, there were reports in the world media of the destruction to property when the Cayahoga River in Cleveland caught fire, and a quote in the *Izaak Walton Magazine* from Dr. Stanford Smith that Lake "Eerie" was dead. Together with the passing of the Clean Water Act in October 1972 to amend the Federal Water Pollution Control Act, the signing of the Great Lakes Water Quality Agreement was seen to represent a significant political response to growing environmental awareness in the United States.

Similarly, the Agreement conferred a benefit on Canada, a country with a numerically much smaller population than the United States, in that the United States agreed to consult and undertake regulatory actions on an equal basis. For example, the numbers of Canadian and United States members on the boards, who advise the International Joint Commission on the effectiveness of activities under the Agreement, are equal. This represented no small concession for the United States, and, in terms of preserving the self interest of Canada, these organizations still represent important consultative instruments in binational relations for avoiding potential irritants concerning transboundary pollution.

Persistent toxic substances and the limitations of the water quality approach

Since the time of the debates between John Van Oosten and Thomas Langlois in the 1940s (Egerton 1985), there has been a controversy about whether pollutants could cause effects at anything but the local level. In the 1970s, however, there was a growing appreciation of the large-scale contamination of the Great Lakes with persistent toxic substances and of their role in causing a variety of effects on Great Lakes wildlife populations. As examples, DDT had been shown to have caused the thinning of eggshells of various bird species nesting throughout the Great Lakes (Anderson and Hickey 1972); the presence of PCBs in fish had caused severe kit mortality in ranch mink (*Mustella vison*) in Michigan and southern Ontario (Aulerich et al. 1973), and it was suspected that the outbreak of chick edema disease in Lake Ontario herring gulls (*Larus argentatus*) was caused by polychlorinated biphenyls, dibenzo-*p*-dioxins, and dibenzofurans (Gilbertson and Fox 1977). The use of some fish stocks in Lake St. Clair and Lake Erie for commercial purposes was being curtailed because of widespread mercury contamination, and sport fish advisories were beginning to be prepared and posted to respond to the findings of human exposures to organochlorine compounds from consumption of Great Lakes fish.

It was in this climate of impending success over the agreements on target loadings for phosphorus and the development of water quality objectives, and in a climate of concern for persistent toxic substances, that the 1972 Great Lakes Water Quality Agreement was renegotiated. The resulting 1978 Agreement had a strong reorientation towards endorsement of policies in the two countries concerning the development and implementation of controls of discharges of toxic substances. Specifically, it included a policy that stated that "the discharge of any or all persistent toxic substances be virtually eliminated." This was a novel policy that reflected the suspicion that the injury to populations of Great Lakes organisms had been caused by exposures to these substances.

At the same time in the mid-1970s, scientists were exploring the limitations of the traditional water quality approach for certain substances, on the basis of the development of water quality objectives and enforcement of standards for effluent discharges. This single-medium approach for some substances, such as compounds of mercury and other trace metals in addition to organochlorine compounds, seemed inadequate. They entered the Great Lakes from a variety of nonpoint sources, such as agriculture and industrial facilities, as well as traditional point sources, such as municipal and industrial discharges; they moved between different environmental media, for example, from the atmosphere to the water and into and out of sediments; and some were concentrated in food chains to levels that were of toxicological significance. The application of technology and regulatory control on sources to one environmental medium sometimes resulted in pollution of another medium. There was a sense that the connectedness of the system was not being reflected in the water quality programs of governments.

Introduction of the Ecosystem Approach

On the basis of these limitations of the single-medium water quality approach, the newly named Great Lakes Science Advisory Board recommended to the International Joint Commission that water quality planners should take a multimedia approach to water pollution. The authors of the report named

© 1997 NRC Canada

this multimedia approach the Ecosystem Approach (International Joint Commission 1978; Dr. J.R. Vallentyne, Department of Fisheries and Oceans, Burlington, Ont., personal communication). The commissioners accepted this recommendation, but warned that the approach should not interfere with progress on the development of water quality objectives, since this was perceived to be a critical part of the bilateral strategy to restore water quality. In the 1978 renegotiation of the Agreement, the ideas contained in this report were incorporated into a statement of purpose of the Agreement as follows: "The purpose of the Parties is to restore and maintain the chemical, physical, and biological integrity of the waters of the Great Lakes Basin Ecosystem," which was defined as "the interacting components of air, land, water and living organisms, including man, within the drainage basin of the St. Lawrence River at or upstream from the point at which this river becomes the international boundary between Canada and the United States." There is an ambiguity implicit in the statement of purpose, since it is uncertain whether the loss of the integrity of the waters had been caused by pollution or by other factors. This ambiguity was almost immediately elaborated by the fisheries scientists into another ecosystem approach with a different meaning (Spangler et al. 1987).

In the early 1970s, fisheries biologists undertook an analysis of variance of the data, presented at the Conference on Salmonid Communities in Oligotrophic Lakes, on the status of salmonid populations in 37 holarctic lakes. From this analysis of variance, they concluded that the effects of several stresses, including eutrophication, overfishing, or the introductions of exotic species, acting on a stock of fish at the same time, could not be distinguished from each other (Loftus and Regier 1972; Regier and Loftus 1972). In essence, the specific cause was thought to be unknowable. They thus advocated an ecosystem approach to the management of all the potential stresses, rather than further research to single out the most important one. At that time, data on persistent toxic substances were deemed to be ambiguous and, although the issue was noted, it was explicitly excluded from the suite of stresses considered in the analysis of variance (Dr. W.J. Christie, Ontario Ministry of Natural Resources, Glenora, Ont., personal communication). This conference and the subsequent International Symposium on Stock Assessment and Yield Prediction (ASPY) (Spangler et al. 1987) seem to have been formative experiences for fisheries researchers and managers and led to a belief that the causes of changes in the status of fish populations were unknowable and that contaminants were probably not an important factor (Regier 1979). This new meaning of the Ecosystem Approach was incorporated into two new and holistic fisheries approaches: A Strategic Plan for Ontario Fisheries and the Strategic Great Lakes Fishery Management Plans (Spangler et al. 1987). A more recent statement reflecting the polythetic or multicausal factors involved in changes in the status of the fisheries has been published by Kitchell (1995). Similarly, Carpenter et al. (1996) and Cooper (1995) have expressed scepticism about the role of pollution by persistent toxic substances in causing effects on fish and wildlife, respectively.

The name Ecosystem Approach connotes ecology and systems theory. Ecology and systems theory are broad integrative sciences lacking parsimony (Dunbar 1980) and were here being applied to what had been the narrow policy field contained in the 1972 Great Lakes Water Quality Agreement. The name may therefore be an unfortunate choice in that the scale of what is implied is much larger than the original subject matter concerning transboundary pollution. These broad integrative sciences and the definition of the Great Lakes basin ecosystem implied no limits on the disciplines or practitioners involved, or on the agenda to be addressed.

There are two purposes to this paper. The first is to explore these differences in interpretation of the Ecosystem Approach between the multimedia approach to water pollution and the fisheries strategy of managing all stresses because the specific causal agent is unknowable. The second is to examine whether there were unintended, as well as the intended, consequences of incorporating, in the 1978 Great Lakes Water Quality Agreement, the broad field of ecology into the narrow policy field of pollution of the boundary waters, and whether this compromised the benefits that both Canada and the United States obtained in creating the 1972 Great Lakes Water Quality Agreement. There are several categories of programs that could have been affected including, for example, design and funding of research and monitoring, preparation and implementation of remedial action plans and lakewide management plans, selection of indicator organisms, and development of objectives. This analysis may be relevant to the possible review and renegotiation of the Agreement in the next few years. It may also be relevant for priority setting at this time of widespread budgetary constraints in both nations. Finally, it may be important in considering recommendations that the Great Lakes Water Quality Agreement should be transformed into a Great Lakes basin ecosystem agreement. For example, this transformation from a focus on water quality to one on maintaining and restoring the Great Lakes basin ecosystem has already occurred in relation to the Canada–Ontario Agreement.

Mandate

It is perhaps useful to start with a review of the basis for the mandate of the 1972 Great Lakes Water Quality Agreement. The Agreement was between the United States and Canadian governments and resulted from negotiations that followed from the successful completion of the 1964 reference to the International Joint Commission concerning the pollution of the lower Great Lakes. The 1964 reference was made pursuant to the Boundary Waters Treaty of 1909, which states in Article IV that the parties "agreed that the waters herein defined as boundary waters and waters flowing across the boundary shall not be polluted on either side to the injury of health or property on the other." The agenda that derives from this mandate is therefore about a limited number of phenomena. This includes pollution of a delimited geographic area and injury to health and property. This is the narrow policy field of the Boundary Waters Treaty of 1909 concerning pollution. The 1972 Great Lakes Water Quality Agreement was a separate document from the treaty, but explicit in the Agreement and implicit in the articles and annexes is the continuing concern "about the impairment of water quality on each side of the boundary to an extent that is causing injury to health and property on the other side." This is an acknowledgement that the one cause of the documented injury is pollution of the boundary waters.

The 1978 introduction of the Ecosystem Approach into this policy field resulted in the expansion of the boundaries around

© 1997 NRC Canada

the mandate of the Agreement during the various renegotiations and amendments. For example, for many scientists and administrators, the term pollution no longer refers only to substances, but includes the introduction of exotic species such as the sea lamprey (*Petromyzon marinus*), zebra mussel (*Dreissena polymorpha*), ruffe (*Gymnocephalus cernuus*), and purple loosestrife (*Lythrum salicaria*). Although the original document (International Joint Commission 1978) refers to introduced species, this is only mentioned in relation to organisms as vectors (vehicles) for transboundary pollution. The geographic area could include not only the waters of the Great Lakes and their sediments, and the air above them, but also the tributaries and all of the wetlands, uplands, farmland, and forests as well as municipalities in the watershed. There is little mention of injury, but there is an extensive elaboration of the concept of risk that now ambiguously seems to include the concept of actual damage as well as potential effects. Similarly, the word property is seldom mentioned, but the term health has taken on a newly expanded meaning from human health, which was probably the original sense in 1909, to the health of fish and wildlife organisms and their populations, and finally to ecosystem health. It is perhaps relevant to ask at this point what is not included under these interpretations of the purview of the Great Lakes Water Quality Agreement, since it would seem to have been extended from the science and administration concerning water quality into consideration of all social, economic, and cultural aspects of all human activities and behaviour, including governance of the Great Lakes basin (Caldwell 1988). Social and economic aspects were mentioned in the original version of the Ecosystem Approach, but only in relation to transboundary pollution.

This process of extending the boundaries of the Great Lakes Water Quality Agreement (Christie 1995), through the various interpretations of the Ecosystem Approach, has served many interests. The availability of relatively larger fiscal resources in the 1970s and 1980s enabled the expansion of research, and thereby the number of issues being addressed by those involved in the implementation of the Great Lakes Water Quality Agreement. Scientists and other professionals from many peripheral disciplines have found a niche in the system, and those from apparently unrelated fields have been able to legitimize their studies and sometimes even usurp or obscure the main focus relating to pollution and water quality. Managers faced with dwindling budgets and a seemingly intractable legacy of toxic chemical wastes in landfill sites and sediments can deflect the spotlight onto issues that are more remediable, such as conservation of wetland habitat and prevention of the introduction of exotic species. Industries that are potentially facing enormous liabilities for injury to human health and to resources from past chemical manufacturing operations can point to the other factors, such as loss of habitat, to obscure and deflect the complaints concerning pollution. The incremental interpretations of the Ecosystem Approach have served many interests, but it is possible that they may not necessarily have served the public interest in relation to transboundary water pollution.

One of the other important consequences of the application of the Ecosystem Approach concerns the statements made by scientists about the cause of the injury. Ecology is a multicausal science, and thus the unicausal statements of chemically induced injury that were arguably efficacious in the past in controlling persistent toxic substances that had caused injury have tended to be rejected (Holling 1992) as simplistic. The assumption is made that observed changes in the status of populations of organisms in the Great Lakes basin, and changes in the incidence of health of organisms, are caused by a large number of factors acting in unison (Kitchell 1995). Similarly, it is assumed that water pollution is only one of these factors and, at most, likely to be of only submarginal importance. For example, there is reputed to be a consensus among Great Lakes fisheries managers that "there is no need to invoke toxic contaminants as directly or indirectly involved in the major dynamics of Great Lakes fish populations" (Carpenter et al. 1996).

Further, the application of chaos theory to the Ecosystem Approach has resulted in an intellectual pessimism among scientists about ever being in a position to make statements concerning the observed injury and the causal relationship to any particular factor, and particularly to pollution by chemicals. Ecologists have applied quantum theory to assert that the lakes have flipped into other equilibrium states and that it will therefore be practically impossible to restore these systems, and thus their administration should be through adaptive management. Questions are posed about the period of historic ecological integrity to be attained, and the impossibility of reaching a pristine state corresponding to pre-Columbian times. An examination of the wording of the Boundary Waters Treaty would indicate that the parties should be trying to restore the water quality to a state that would correspond to the period of about 1930, which was the most likely time when manufacture and use of persistent toxic substances rose and the injury to health and property started to occur, although not to be detected or understood. Thus, discussions about what would constitute a pristine state are largely irrelevant in relation to the implementation of the Treaty and the Agreement.

Consequences for implementation of the Great Lakes Water Quality Agreement

There is no doubt that the two countries have both benefitted from the burgeoning economic situation in North America that has, during this century, produced not only enormous wealth, but also the severity of the pollution of the Great Lakes by municipal and industrial wastes and agricultural operations. The United States has invested heavily in the Great Lakes region in the development of an extensive infrastructure for research and development in chemical manufacturing and in production for export as well as the domestic market. In contrast, Canada has been a branch plant operation, in terms of chemical manufacturing and research, producing bulk quantities of staple products for the national market. Thus, the potential for injury from the chemical manufacturing operations located in the United States is much greater than from those in Canada, from the standpoint not only of the volume of the chemicals produced, but also of the number and diversity of compounds entering commerce (Leonardelli 1995). Has there been injury from these operations, and particularly, has there been injury to health and property caused by transboundary pollution from the United States and Canada? If the Agreement already contains the goals for cleanup and management, do we need to know the detail of the relationship between the injury to health and property and the specific causes? Or is it

© 1997 NRC Canada

naive to expect regulatory officials to undertake new and costly remedial actions without a proven cause and effect (Great Lakes Water Quality Board 1993)?

Despite the use of the words "cause" or "causing" in the text of the 1972 and 1978 agreements, in the articles and in the annexes, there is a strong aversion among water quality scientists to involving themselves in making statements about the injury that occurred and its causes. For example and with italics added, the 1972 Agreement states that the parties were "[S]eriously concerned about the grave deterioration of water quality on each side of the boundary to an extent that is *causing* injury to health and property on the other side." Annex 2 of the 1978 Agreement is concerned with the preparation of remedial action plans and lakewide management plans. The definitions of an area of concern and of a critical pollutant both contain references to substances that are at levels that cause impairments of beneficial uses. The plans that have so far been submitted have been noticeably lacking in causal statements. Whether this is the result of a lack of knowledge of the literature or simply professional cautiousness among scientists and engineers about making these kinds of statements, or the reluctance of their institutions to embroil themselves in litigation that might lead to a finding of liability, are moot points, but the absence from the plans of definitive statements of the pollutant-induced injury to health and property has hindered the production of scientifically defensible and focussed plans for effective remedial actions. Unless scientists and managers are prepared to make these statements about the specific injury and the specific chemical causes, it is unlikely that there will be further tangible progress in restoring the populations of organisms injured by releases of and exposures to persistent toxic substances. It seems that only preventive research and statements have become acceptable, even though the rule of law continues to require definitive evidence of injury, of the specific cause, and of those culpable.

The situation is more serious with respect to statements made by ecologists using the Ecosystem Approach. In the negotiations that led to the 1987 Protocol that amended the 1978 Great Lakes Water Quality Agreement, there was inclusion, in the supplement to Annex 2 concerning the preparation and implementation of remedial action plans and lakewide management plans, of an explicit transformation from the unicausal to a multicausal assumption about injury. The ambiguity contained in the stated purpose of the 1978 Agreement was clarified in the definition of the "Impairment of beneficial use(s)". The changes "in the chemical, physical and biological integrity of the Great Lakes System" were no longer the effects of water pollution, but became the causes of the 14 use impairments. In fact, following systems theory of the interconnectedness of everything, any reference to water in the definition was omitted. The inclusion of the "degradation of aesthetics" and the "loss of fish and wildlife habitat" resulted in a further shift of the focus of the Agreement away from injury by pollution towards an emphasis on a wide array of physical, chemical, biological, and even socioeconomic factors. The consequence has been that the agenda of those organizations involved in the implementation of the Great Lakes Water Quality Agreement has included a series of new items, most of which, although they are all doubtlessly important topics, are only marginally relevant to the originally stated purpose of the 1972 Agreement and of the Boundary Waters Treaty concerning pollution of the boundary waters.

These items, for example, include fisheries management, the introduction of exotic species, wetlands and habitat policy and practice, biodiversity, socioeconomic activity, governance, globalization, and sustainable development. The inclusion of these open-ended items in the agenda for the implementation of the Great Lakes Water Quality Agreement is justified on the basis that this is a requirement in implementing an Ecosystem Approach based on systems theory. But their inclusion has had a profound effect on the interpretation of the mandate. For example, in many jurisdictions, the preparation of the remedial action plans is frequently the only local planning process with the active participation of the public. From a bilateral perspective, it has sometimes been difficult for observers in other jurisdictions to view decisions, based on these new interpretations of the Ecosystem Approach, as legitimate remedial actions pursuant to the Great Lakes Water Quality Agreement. For example, environmental managers in other jurisdictions have looked askance at decisions to build parks, bicycle trails, information booths, artificial islands for breeding birds, and fish barriers to restore wetlands, rather than remove contaminated sediments and remediate leaking chemical landfill sites that continue to release persistent toxic substances into the boundary waters.

Development of Great Lakes forensic toxicology

To overcome these limitations of the water quality approach and the multicausal interpretation of the Ecosystem Approach, and the scepticism of the scientific and regulatory communities concerning the alleged injury, a loose coalition of scientists, centred on the International Joint Commission and involved in the health of fish, wildlife, and humans, have, since about 1988, developed a forensic toxicology approach. The forensic approach assumes that the number of factors that caused the observed injury to health and property is small, that the factors are knowable, and that one of the significant factors was and continues to be persistent toxic substances. The forensic approach relies on epidemiological criteria to reject spurious factors and to affirm probable causal relationships on which definitive statements can be made to regulatory officials. This has resulted in significant progress in the quality of the statements that these scientists have been prepared to make, both in the scientific literature and to the courts and regulatory authorities. This group of scientists comprises mainly field biologists, biochemists, and analytical chemists associated with certain fish and wildlife research organizations.

This initiative to make more definitive statements has been undertaken by a group of scientists presenting their work in a series of Cause–Effect Linkages Workshops sponsored variously by the International Joint Commission (Gilbertson and Schneider 1991), the Michigan Audubon Society (Gilbertson and Schneider 1993), and the International Association for Great Lakes Research (Gilbertson 1996). These workshops have resulted in the development of a forensic approach to link injury to the health of fish, wildlife, and human populations to specific pollutants in the Great Lakes. The challenge for scientists has been to reduce their uncertainties about the evidence concerning the injury to populations of organisms in the

© 1997 NRC Canada

Great Lakes basin and about the causal relationship with exposures of those populations of organisms to specific persistent toxic substances. This is not a new challenge for scientists involved in diagnosing diseases and deducing their causes. During the past 150 years, epidemiologists and clinicians have developed a series of epidemiological criteria for integrating and judging the evidence, and for rejecting the putative causes that are clearly spurious and tentatively affirming the putative causes that seem to fit the evidence (Susser 1986). Similar criteria have been developed for the physical sciences (Popper 1962, 1968).

The initial advance in developing the forensic approach in the Great Lakes basin came from the distribution of this methodology to other scientists (Fox 1991) and its application to the existing evidence relating to the case studies of injury thought to be related to exposures to persistent toxic substances. Unfortunately, the methodology and these case studies have been ignored by most of the scientific and regulatory authorities involved in the implementation of the Great Lakes Water Quality Agreement as not an authentic part of the agenda. Instead, the emphasis has traditionally been on the experimental determination of the toxicological characteristics of newly detected compounds and the derivation of water quality objectives and on statements about the potential effects based on risk assessment. It would seem that Canadian and United States scientists and environmental managers, in making these statements about potential effects, have undermined the seriousness of their complaint when actual injury to individual organisms and to populations had occurred as a result of transboundary pollution. Thus, these scientifically responsible statements may not have been socially responsible, particularly among government researchers and managers, because they did not lead to an appreciation among regulatory officials of the severity of the damage, the certainty of the causal evidence, and the scale of the remedial response that would be required.

The most convincing case studies concern the effects on fish-eating birds. Early in the 1960s, Keith (1966) and Ludwig and Tomoff (1969) estimated the reproductive success of herring gulls in colonies on Lake Michigan islands. They related the reproductive failure to exposures to DDT and metabolites and to dieldrin. Sprunt et al. (1973) reported the reproductive failure and population declines of bald eagles (*Haliaeetus leucocephalus*) on the North American continent and showed that the most severe effects and highest concentrations of organochlorine compounds occurred in the relict population in the Great Lakes basin. In the mid-1960s, there was a serious outbreak of kit mortality in ranch mink in farms in Michigan and Ontario that was traced to PCB contamination of Great Lakes fish used as feed (Aulerich et al. 1973). In the early 1970s, the findings of severe embryo and chick mortality and of deformities in Lake Ontario colonies of several species of fish-eating birds led to a controversial diagnosis of chick edema disease and, in the early 1980s, to the eventual detection of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin in eggs of herring gulls (Gilbertson et al. 1991). Similar observations were made in the early 1980s on a colony of Forster's terns (*Sterna forsteri*) in Green Bay (Kubiak et al. 1989), Wisconsin, and in the late 1980s on Caspian terns (*Hydroprogne caspia*) and double-crested cormorants (*Phalacrocorax auritus*) (Tillitt et al. 1992; Ludwig et al. 1996), and these were mainly attributed to exposures to high levels of PCBs in the fish. Similar kinds of observations were made in the 1980s on embryos and hatchlings of snapping turtles (*Chelydra s. serpentina*) living and nesting on the shorelines of the lower Great Lakes (Bishop et al. 1991). The pathological and reproductive effects of Great Lakes pollutants were even documented in beluga whales (*Delphinapterus leucas*) in the Gulf of St. Lawrence that had become contaminated through ingestion of American eels (*Anguilla rostrata*) migrating from Lake Ontario to the Sargasso Sea (Béland et al. 1993). These case studies would seem to represent a significant response to the originally identified need that was named the Ecosystem Approach (International Joint Commission 1978).

With some notable exceptions (Mac and Edsall 1991), there is a noticeable lack of case studies relating effects in Great Lakes fish to exposures to chemicals. The assumption among personnel in fisheries agencies is that fish are relatively insensitive to persistent toxic substances in general and organochlorine compounds in particular and that exposures of fish to these substances in the offshore, pelagic regions of the lakes are low. The results of several efforts to correlate the incidence of embryo or fry mortality in various salmonids to the levels of pollutants have been inconclusive (Ankley et al. 1991; Smith et al. 1994), but the experimental demonstration of the embryotoxicity of extracts of rainbow trout (*Oncorhynchus mykiss*) from Lake Ontario to medaka (*Oryzias latipes*) is suggestive of a possible role of bioaccumulated pollutants (Harris et al. 1994).

New information from a variety of toxicological studies and from an exposure analysis based on radiodated sediment cores from Lake Ontario has been used to construct a retrospective risk assessment (Dr. Philip Cook, U.S. Environmental Protection Agency, Duluth, Minn., personal communication). The results indicate that the levels of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin were sufficiently high in the 1940s to have precluded the hatchability of any Lake Ontario lake trout (*Salvelinus namaycush*) eggs. The indigenous lake trout in Lake Ontario were extinct by the 1960s. Similar radiodated sediment cores show that organochlorine pollutants were in other Great Lakes prior to the extinctions of their lake trout populations. It has been widely assumed that these and other extinctions were successional changes (Smith 1972; Christie et al. 1987) attributable to eutrophication, the depredations of the sea lamprey, and overfishing, but not to chemical pollution (Eshenroder et al. 1984; Kitchell 1995; Carpenter et al. 1996). These assumptions must now be reevaluated in relation to all fish extinctions in the Great Lakes during this century, and particularly of those species, such as deepwater sculpins (*Myoxocephalus quadricornis*) and burbot (*Lota lota*), that lived in the deep profundal areas. Persistent toxic substances were adsorbed on to fine materials that were selectively resorted and transported to these deepwater habitats (Howdeshell and Hites 1996).

If the thesis proposed by Dr. Cook eventually becomes accepted, it will have exemplified the length of time between the occurrence of injury to fish populations caused by persistent toxic substances and the perception of that injury and the length of time to undertake relevant research and subsequent acceptance of a scientifically defensible explanation. Similarly, it will throw fisheries toxicology research into an intellectual crisis as the existing body of knowledge is dismantled and reconstructed around the new paradigm (Kuhn 1962). A

© 1997 NRC Canada

corollary of the doubt that persistent toxic substances had any effect on the status of fish populations (Carpenter et al. 1996) is that there has been little or no support from fisheries agencies or environmental agencies to implement the policy concerning the virtual elimination of discharges of persistent toxic substances to the Great Lakes. In addition, United States fisheries agencies with burgeoning demands from sport fishermen could manage around the problems of reproductive failure in salmonids, and the uncertainties about the causes, by stocking lake trout and the newly imported Pacific salmon from uncontaminated brood stock in hatcheries. The scientific debate has been further obscured by the emotional politicization of the issue of contaminants and fish by fisheries interests and agencies.

The lack of interest among the fisheries agencies was paralleled by that of the authorities responsible for human health. An epidemiological study of the infants of mothers who had eaten large amounts of fish from Lake Michigan was initiated in the late 1970s (Fein et al. 1984). Previous studies had shown that the levels of contaminants in the blood of people who ate Great Lakes fish were higher than those who did not eat Great Lakes fish (Humphrey 1983). Although there did not appear to be any effect on the people themselves, there was a growing concern for the potential for effects to occur in their offspring. Infants of mothers who had eaten about a pound per month of Lake Michigan fish for 6 years prior to pregnancy were smaller at birth and had poorer psychomotor development (Fein et al. 1984). These findings were correlated with the levels of PCB in the blood in the umbilical cord sampled at birth. Subsequent testing at 7 months and at 4 years showed that the growth retardation was persistent and that the children of the mothers who consumed larger quantities of Lake Michigan fish had poorer cognitive development and short-term memory (reviewed in Jacobson 1994). A subsequent study by other researchers (Dar et al. 1992) and an epidemiological critique (Paneth 1991) tended to blunt these findings. Infants of Green Bay, Wisconsin, mothers who had eaten relatively small amounts of fish compared with mothers in the the previous study, prior to pregnancy, were larger than those who had eaten no fish and their size correlated with the maternal PCB serum levels (Dar et al. 1992). Further testing of the Michigan cohort by the Jacobsons at 11 years showed that the children of the mothers who had eaten the most Lake Michigan fish had IQ scores that were six points lower than those of the reference group (Jacobson and Jacobson 1996), indicating that the chemically induced neurological deficits detected at birth were persistent.

The health authorities in both countries belatedly secured funding in the late 1980s and have undertaken or funded a series of investigations that are beginning to delineate the extent of the dangers, particularly to native people, sports anglers, pregnant and nursing mothers, fetuses, nursing infants and children, the urban poor, and the elderly (DeRosa et al. 1995). The neonatal findings of the Jacobsons were eventually replicated in another epidemiological study undertaken on another cohort of infants of mothers who had consumed fish from Lake Ontario (Lonky et al. 1996). They did not find the changes in gross development found by the Jacobsons in their cohort, perhaps because the levels of pollutants had declined in the 1980s. However, they did confirm the findings of poorer psychomotor development at birth. In addition, on the basis of experimental findings on rats (Daly 1993), they undertook an assessment of the response to mild frustration and found that the infants whose mothers had eaten the larger amounts of Lake Ontario fish habituated to the frustrating stimuli less than the infants of the non-fish eaters (Lonky et al. 1996).

It would seem that the institutional arrangements for communicating these findings and for responding to these various pieces of evidence have been ineffective. The failure of the fisheries and wildlife authorities to present their case studies to the requisite national and international authorities poses the question of the proper role and functioning of government and of the officials charged with protection of the natural resources. The failure of the health authorities in both countries to respond promptly to the findings in wildlife and to the initial epidemiological findings in neonates, other than to issue advisories, reflected the politics of the late 1970s and the 1980s. Funding for investigations of the human health implications of contaminants in Great Lakes fish and wildlife was only made available in the late 1980s and early 1990s, after tens of thousands of infants over several decades had been exposed in utero to pollutants from maternal consumption of contaminated fish.

Consequences of the forensic approach

This forensic approach has yielded two products. First, it has resulted in a series of case studies that now constitutes the specific complaint, by scientists of both nations, that pollution by specific persistent toxic substances of the waters of the Great Lakes has resulted in observed injury to human health and to wildlife health and to the use of fish and wildlife as resources. At this stage the connection between exposures to persistent toxic substances and effects on fish populations still remains contentious. This complaint poses the question to the pollution control authorities in both countries concerning their appropriate response to this evidence of continuing injury, in violation of the Boundary Waters Treaty and the Great Lakes Water Quality Agreement.

The second product of the forensic approach has been to provide a toxicological interpretation of the Great Lakes Water Quality Agreement rather than the successional viewpoint espoused by the ecologists and fisheries managers. In the context of forestry management, Holling and Meffe (1996) have provided an analogue. They described problems of natural resources management in the natural world as being complex, nonlinear, and poorly understood, in contrast with the problems of controlling human activities that tend to be problems that are well bounded, clearly defined, relatively simple, and generally linear with respect to cause and effect. The new forensic evidence poses the question as to whether the problems of pollution of the boundary waters, particularly by persistent toxic substances, are really complex, unbounded, and poorly understood as the ecologists would have us believe. The corollary that follows is whether the ecologists could have unwittingly led the Great Lakes scientific and regulatory community into a quagmire of complexity (Kay and Schneider 1994) that simultaneously precluded the effective delivery of the forensic complaint relating to chemically induced injury to health and property.

© 1997 NRC Canada

Bilateral responses and the advice to move on to other agenda

Canada and the United States are not only neighbours and allies but also trading partners, and pollution, particularly transboundary pollution, is looked upon as a potential irritant to these harmonious economic relationships between the two nations. The signing of the Boundary Waters Treaty in 1909 and of the various versions of Great Lakes Water Quality Agreement in 1972 and 1978 has been the diplomatic response of the nations to these potential irritants. The International Joint Commission became an important bilateral forum, through the Great Lakes Water Quality Board and the Great Lakes Science Advisory Board, established under the Agreement, for discussion of the evidence, development of water quality objectives, and development of remedial plans. These arrangements worked fairly well during the 1970s and 1980s, and the Commission developed a reputation for enabling bilateral consultation and interjurisdictional cooperation and for the quality of its work through its boards and associated secretariat. During the 1987 review and amendment of the 1978 Agreement, the parties removed these functions and responsibilities from the International Joint Commission, partly to better control the developing program on remedial action plans and lakewide management plans and partly to ensure that they were acknowledged for producing the results of the surveillance and monitoring programs that were traditionally reported by the Great Lakes Water Quality Board to the International Joint Commission. The parties have, however, found it difficult to establish satisfactory mechanisms for implementation of the Agreement in the absence of any permanent bilateral secretariat set up for this purpose outside the International Joint Commission.

The question now posed is whether the various research and regulatory programs have enabled the two governments to effectively deliver their policy on the virtual elimination of discharges of persistent toxic substances. There has been a significant ambiguity about objective measurement of the delivery of this policy. The parties, in reporting on activities under the Agreement, refer to the degree of compliance with water quality objectives. Implicit in this traditional water quality approach is the assumption that if there is compliance with the established objectives, there is no legal injury. However, since the original complaint is stated in terms of injury to health and property, perhaps the appropriate measures should be reported in terms of whether the biological injury is still occurring despite compliance with water quality objectives, significant reductions in the releases of persistent toxic substances, and declines in their concentrations in the environment and in indicator organisms.

There has been a significant discussion in the recent ecological literature concerning the improvement in the status of many of the organisms previously affected by persistent toxic substances (Cooper 1995; Kitchell 1995; Smith 1995; Carpenter et al. 1996). The argument is made that the control measures have been so successful that it is now time to move on to other Great Lakes ecosystemic issues such as the introduction of exotic species, wetlands and habitat restoration, biodiversity, and sustainable development, since the focus of the Ecosystem Approach, first and foremost, is on the human behaviour that generates environmental problems (Dr. J.R. Vallentyne, Department of Fisheries and Oceans, Burlington, Ont., personal communication). For example, the preparation, by the parties to the Agreement, of the lakewide management plan for Lake Erie, which is meant to be an analysis of the injury to the uses of resources caused by critical pollutants, initially tended towards becoming a fisheries management plan, in which the issue of pollutants was relegated to a submarginal concern. An alternative interpretation (Ludwig 1996) of the urge to move on is that the problems of remediation of the sources and sinks of persistent toxic substances are being perceived by scientists, managers, and regulatory officials to be so intractable that the policy of virtual elimination of the discharges of persistent toxic substances is being abandoned. There are two objections to this urge to move on to other topics.

First, there has been almost no progress in the past 10 years by the parties to the Great Lakes Water Quality Agreement in selecting reliable indicator organisms and in collecting and reporting the status of those indicator organisms in relation to the policy of virtual elimination. In fact, there is so little discussion or agreement within the scientific community, and particularly within senior management of environmental agencies, about what would constitute a reliable indicator that it is still premature to declare that the policy on virtual elimination of discharges of persistent toxic substances has been satisfactorily delivered. This lack of progress in the selection of indicator organisms and in the development of objectives has been partly due to another ambiguity, introduced into the 1987 Protocol that amended the 1978 Agreement, of incorporating the Ecosystem Approach. The new text now mentioned the development of "ecosystem indicators." It was left as a moot point as to whether these were to be indicators that reflected the state of the Great Lakes, specifically in relation to persistent toxic substances and other pollutants, or whether they were to indicate the state of the entire Great Lakes basin ecosystem regardless of whether the factor(s) that had caused the changes was known or unknown, man-made or natural, or related to specific pollutants or to multiple other factors. It would seem that in the past decade, multicausal statements have become the only ones that most scientists and administrators would regard as correct. The notable exception to this process was the development of water quality guidance for the Great Lakes by the U.S. Environmental Protection Agency (1995), which, almost unilaterally, undertook the novel approach of establishing water quality criteria for fish-eating wildlife and for humans, specifically for the persistent toxic substances known to have caused injury.

The second objection to moving on to other topics relates to the letter and spirit of Article IV of the Boundary Waters Treaty as the basis of the Great Lakes Water Quality Agreement. In the last century and the early part of this century, the pollution of boundary waters with domestic and municipal sewage was recognized as a serious source of microbiological disease agents that could cause human sickness and death in human communities in the other country. Similarly, transboundary pollution with industrial wastes was known to pose serious threats to resources on the other side. In this century, there has been a growing appreciation of the injury to health and property from exposures to a wide variety of persistent toxic substances released to the boundary waters from chemical manufacturing industries and widespread domestic,

© 1997 NRC Canada

industrial, and agricultural use of chemicals. At the time of the 1909 signing of the Boundary Waters Treaty, it had long been recognized that transboundary pollution was a significant potential bilateral irritant to harmonious diplomatic relations between the two countries.

Long after the Boundary Waters Treaty had laid out the general agreement against transboundary pollution, the International Law Association (1967, 1983) set out the principles and obligations of states that share a drainage basin, including notification of the other nation of activities that might be or have been deleterious to the interests of that other nation. Similarly, there was a general prohibition on the release of substances that were generally regarded as ultrahazardous (Bourne 1986). Transboundary pollution of shared water bodies is now universally recognized as a serious potential irritant between neighbouring states. The question now posed to those advocating that the agenda should move on to other, doubtlessly important, topics is whether their other issues, such as the introduction of exotic species, wetland and habitat degradation, and depletion of fisheries, pose such a serious threat to national sovereignty and bilateral relations that they warrant displacing transboundary pollution by chemicals and pathogenic microorganisms as the proper focus of the Great Lakes Water Quality Agreement. Or should the Agreement be used to manage all human activities and resources in the Great Lakes basin ecosystem? This is the legacy of ambiguity left by the negotiators of the 1978 Great Lakes Water Quality Agreement and of its amendment by protocol in 1987.

Over the past 20 years, there have been several expressions of the need for an ecosystem approach to management of the Great Lakes and human activities within the basin and even ecosystem management using the Great Lakes Water Quality Agreement (National Research Council of the United States and the Royal Society of Canada 1985; Francis and Regier 1995; Koonce 1995). There are, however, institutional mechanisms other than the Great Lakes Water Quality Agreement for addressing these other issues. For example, some of these issues, such as wetland and habitat degradation, are of mainly local concern and should be addressed through the local or regional planning authorities and not threaten the diminishing resources allocated in the 1990s to the implementation of the policies concerning lakewide pollution under the Great Lakes Water Quality Agreement. There are such national institutions as the Great Lakes Commission and the respective Coast Guards and international institutions such as the Great Lakes Fishery Commission for addressing the threats posed by the introduction of exotic species. Similarly, the Great Lakes Fishery Commission is the proper place for addressing the complex issues of fisheries management, including biodiversity and wetland habitat restoration. The Migratory Bird Convention and the North American Waterfowl Management Plan are suitable institutions for addressing biodiversity, endangered species, and the protection and restoration of waterfowl habitat. None of these issues, however, would seem to pose the same scale of threat to national sovereignty and bilateral interests as does transboundary pollution, and thus, none would seem to warrant the degree of attention that has been and should continue to be paid to assessing and controlling releases of pollution that injure fish, wildlife, and human health under the Great Lakes Water Quality Agreement. In these times of declining fiscal resources, there is the need for a clarification of the respective roles and responsibilities of these national and bilateral organizations and for improved consultative mechanisms among them rather than attempts to appropriate each other's agenda. If necessary, the United States and the Canadian governments can develop separate and new agreements on these other bilateral issues and devise more effective mechanisms for their bilateral coordination.

At least five recent scientific developments indicate that the hazards posed by persistent toxic substances, and particularly organochlorine compounds, are more serious than has been generally suspected. These developments contradict the advice that the focus of the Great Lakes Water Quality Agreement should be changed from pollution of the boundary waters.

First, there has been a growing body of evidence that the more toxic, non-*ortho*-substituted congeners of PCBs are selectively bioaccumulated in food webs compared with the *ortho*-substituted PCBs (Kubiak et al. 1989; Smith et al. 1990; Jones et al. 1993; Yamashita et al. 1993) that tend to be sequestered into the sediments (Burkhard et al. 1985).

Second, there is evidence that the presence of the *ortho*-substituted PCBs were antagonistic to the non-*ortho*-substituted PCBs and thus protected the terminal predators from the full expression of the effects (Bannister et al. 1987; Silkworth et al. 1993). The levels of the PCBs and other organochlorine compounds with a similar mode of action have declined in the past 20 years, but there are indications that the actual incidence of effects has not declined at a proportional rate (Ludwig et al. 1996).

The third factor is the new appreciation of the subtle, but devastating effects of these chemicals, particularly on the endocrine systems of all vertebrates, including humans (Colborn and Clement 1992). For a long time, there has been an appreciation of the role of these compounds in the production of a variety of structural deformities in developing embryos of a variety of organisms in the Great Lakes basin (Gilbertson et al. 1976; Bishop et al. 1991; Fox et al. 1991). Only recently has an understanding emerged of the manner in which these compounds interfere with endocrine systems and cause functional deficits of the developing neurological and immune systems.

There are the anomalous dose–response curves for several endocrine systems in which effects at high concentrations are lower than at intermediate concentrations (vom Saal et al. 1995). From a mechanistic standpoint, it seems that at higher concentrations the endocrine systems downregulate. Therefore, with declining levels of pollution, there may be a greater manifestation of effects.

Finally, there is a new appreciation of the scale of the global redistribution of persistent toxic substances and of the selective deposition in colder areas of the world including the Great Lakes and the Arctic (Simonich and Hites 1995) and a growing controversy as to whether concentrations of persistent toxic substances are continuing to decline or whether they have reached a new equilibrium (Smith 1995; Stow et al. 1995).

The implications of all of these factors are that present concentrations of persistent toxic substances such as PCBs, despite 20 years of regulation prohibiting manufacture and certain uses, continue to cause an elevated incidence of effects and thus pose continuing irritants to bilateral relations. They

© 1997 NRC Canada

underscore the prescience of the negotiators of the 1978 Great Lakes Water Quality Agreement when they formulated the policy on virtual elimination of discharges of persistent toxic substances, and the forethought of the International Law Association in formulating the general principle concerning the prohibition of the discharge of ultrahazardous substances into water bodies shared between nations. More particularly, they pose the question whether or not further regulatory and remedial actions are warranted. They indicate that the agenda of the Great Lakes Water Quality Agreement should not, following systems theory, move on to other ecosystemic issues.

Sources of advice in renegotiating the Great Lakes Water Quality Agreement

Much of the advice to governments and to the International Joint Commission on the biological significance of persistent toxic substances has come from fisheries scientists and managers from agencies and academic departments. The water quality objectives and criteria that have been developed over the past 30 years have mostly been designed by fisheries scientists primarily to protect fish. It has long been suspected that these were inadequate to protect and restore certain avian and mammalian wildlife populations (Ludwig et al. 1993). The recent publication of the new criteria for water quality guidance for the Great Lakes indicates that the original criteria were several orders of magnitude too high (U.S. Environmental Protection Agency 1995) to protect or restore wildlife and to protect human health. The new evidence (Dr. Philip Cook, personal communication) of the critical role of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin, 55 years ago, in the extinction of the Lake Ontario lake trout is a significant challenge to the consensus (Carpenter et al. 1996) within the fisheries scientific community of the unimportance of organochlorine chemicals to the status of populations of Great Lakes fish. The date for negotiation of a new Great Lakes Water Quality Agreement may be sooner than the period of time required for resolution of this challenge to the beliefs within the Great Lakes fisheries agencies.

These factors are significant for the discussion of the possibility of renaming the Great Lakes Water Quality Agreement the Great Lakes Basin Ecosystem Agreement and reframing the content of the document to focus on habitat restoration and prevention of the introduction of exotic species, instead of the focus on water quality. The governments can, of course, negotiate whatever agreement(s) they feel is (are) necessary for them to manage the Great Lakes basin ecosystem and all human activities within the watershed basin, but such an ecosystem agreement, based on the successional interpretation of the ecologists and the concept that specific causes of observed effects are unknowable, would do nothing to further restore the biological integrity of the waters of the Great Lakes basin from the continuing incidence of structural and functional teratogenic effects caused by the releases of persistent toxic substances during the past 100 years. Similarly, it would do little to protect future generations of Great Lakes citizens from exposures to environmental teratogens from future releases of persistent toxic substances from past inadequate disposal practices.

Conclusions

The past 40 years of progress on water quality in the Great Lakes have been achieved through bilateral cooperation in research, monitoring, and implementation of regulatory controls. However, during that time, at least three distinct and mutually uncomprehending solitudes have evolved in relation to three or more different and sometimes antithetical approaches to the interpretations of the Great Lakes Water Quality Agreement. These comprise the water quality approach, at least two ecosystem approaches, and the forensic approach. Their contending claims have puzzled managers responsible for implementation of the Great Lakes Water Quality Agreement, and there is now a need for senior management of health, resources, and environmental agencies to reconcile the differences and integrate the approaches.

The water chemistry approach of the 1960s and early 1970s, practised by water chemists and aquatic toxicologists and implemented by civil engineers, was useful for reducing general levels of gross pollution through statements concerning the potential effects of pollutants at particular concentrations. The advantage of the water chemistry approach is that it can be used to predict the likely effects of a particular course of action if certain objectives or standards are exceeded. Thus, it has been used extensively to prevent injury by pollutants as well as for restoration of beneficial uses of water. The approach was limited in its successful application to multimedia sources, distribution, and fate of persistent toxic substances that had caused actual effects on organisms dependent on Great Lakes food chains. In responding to these limitations that were identified in the late 1970s, limnologists, fisheries biologists, and ecologists advocated an ecosystem approach.

The name the Ecosystem Approach poses several problems for scientists and administrators involved in implementing the Great Lakes Water Quality Agreement. First, ecology is a multicausal science, and ecologists tend to include and accumulate putative causal agents rather than reject them. Thus, the legitimate agenda expanded to include many issues that had formerly been quite peripheral to the quality of the waters of the Great Lakes, and persistent toxic substances were treated as just another putative factor. Second, ecology is a broad integrative science and it was being applied to what was originally a narrow policy field derived mainly from a mandate under Article IV of the Boundary Waters Treaty and relating to injury to health and property caused by transboundary water pollution. The consequence was to broaden the geographic ambit of the 1978 Great Lakes Water Quality Agreement to include the entire Great Lakes ecosystem, which could correspond to the watershed and the airshed and everything that lived in it. Third, fisheries scientists have interpreted the Ecosystem Approach to mean that all potential stresses on fish populations should be managed because the specific stress affecting a particular fish population is unknowable.

There is reputed to be a consensus within the fisheries community that the status of populations of Great Lakes fish was not affected by persistent toxic substances except locally in rivers, bays, and harbours. Thus, fisheries scientists have been unable to give definitive statements to officials concerning the injury to health and property caused by persistent toxic substances on which specific further regulatory action could be undertaken. In responding to these limitations and to demands by regulatory officials for proof of a causal relationship between injury and persistent toxic substances, wildlife biolo-

© 1997 NRC Canada

gists, in the late 1980s, advocated a forensic approach. Wildlife biologists, using a forensic approach based on epidemiological principles, have successfully related effects observed in Great Lakes wildlife populations to the toxicological action of specific pollutants or classes of pollutants. The forensic approach assumes that the number of factors that caused the observed injury to health and property is small, that the factors are knowable, and that one of the significant factors was and continues to be persistent toxic substances. The forensic approach relies on epidemiological criteria to reject spurious factors and to affirm probable causal relationships on which definitive statements can be made to regulatory officials.

It is essential in these days of declining budgets in both countries that any forthcoming renegotiation of the Great Lakes Water Quality Agreement should refocus the content onto the core activities, which are to maintain and restore water quality in the boundary waters. This focus must include scrupulous attention, first, to scientific evidence of the chemically induced injury to health and property rather than reliance on risk assessment of potential effects and, second, to the development of water quality objectives appropriate for restoration and protection of populations of critically exposed organisms in the Great Lakes basin rather than dependence on extrapolations from laboratory experiments.

In addition to the implementation of the core activities on water quality, there must be adequate provision for other Great Lakes issues, such as physical habitat restoration, prevention of the introduction of exotic species, fisheries resource allocation, protection and management, governance of the Great Lakes basin, and sustainable development, to be effectively addressed through improved institutional arrangements. It is only through these means that Canadians and Americans will enable themselves to protect their health and interests from injury caused by transboundary pollution. The benefit for the United States of this position in the forthcoming renegotiation would be to help refocus attention on the remediation of the critical landfill sites and other sources of substances that are causing the injury. For Canada to negotiate from any other position would be to relinquish the valuable benefits that accrued from the signing of the Boundary Waters Treaty and the original Great Lakes Water Quality Agreement 25 years ago.

Acknowledgements

In preparing this Perspective, I have been aided by discussions with several scientists and managers who have been involved in various aspects of the Great Lakes Water Quality Agreement. The interpretation that I have placed on the material seems to be controversial and should not necessarily be attributed to any of those named below. I should, however, still like to thank the following: Dr. E. Addison, Mr. B.L. Bandurski, Dr. P. Béland, Mr. P.C. Boyer, Dr. M.P. Bratzel, Dr. P.E. Bertram, Dr. W.J. Christie, Dr. P.M. Cook, Dr. J.M. Cooley, Dr. D.P. Dodge, Dr. J.L. Fisher, Mr. K. Fuller, Dr. A.L. Hamilton, Mr. F.J. Luckey, Dr. J.P. Ludwig, Dr. H.A. Regier, Dr. H. Shear, Ms. D. Stewart, Dr. N. Thomas, Dr. J.R. Vallentyne, and two anonymous reviewers.

References

Anderson, D., and Hickey, J.J. 1972. Eggshell changes in certain North American birds. *In* Proceedings of the XVth International Ornithological Congress. *Edited by* K.H. Voous. E.J. Brill, Leiden. pp. 514–540.

Ankley, G.T., Tillitt, D.E., Giesy, J.P., Jones, P.D., and Verbrugge, D.A. 1991. Bioassay-derived 2,3,7,8-tetrachlorodibenzo-*p*-dioxin equivalents in PCB-containing extracts from flesh and eggs of Lake Michigan chinook salmon (*Oncorhynchus tshawytscha*) and possible implications for reproduction. Can. J. Fish. Aquat. Sci. **48**: 1685–1690.

Aulerich, R.J., Ringer, R.K., and Iwamoto, S. 1973. Reproductive failure and mortality in mink fed on Great Lakes fish. J. Reprod. Fertil. **19**(Suppl.): 365–376.

Bannister, R.D., Davis, D., Zacharewski, T., Tizard, I., and Safe, S. 1987. Aroclor 1254 as a 2,3,7,8-tetrachlorodibenzo-*p*-dioxin antagonist: effects on enzyme induction and immunotoxicity. Toxicology, **46**: 29–42.

Béland, P., DeGuise, S., Girard, C., Lagace, A., Martineau, D., Michaud, R., Muir, D.C.G., Norstrom, R., Pelletier, E., Ray, S., and Shugart, L.R. 1993. Toxic compounds and health and reproductive effects in St. Lawrence beluga whales. J. Great Lakes Res. **19**: 766–775.

Bishop, C.A., Brooks, R.J., Carey, J.H., Ng, P., Norstrom, R.J., and Lean, D.R.S. 1991. The case for a cause–effect linkage between environmental contamination and development in eggs of the common snapping turtle (*Chelydras s. serpentina*) from Ontario, Canada. J. Toxicol. Environ. Health, **33**: 521–547.

Bourne, C.B. 1986. International law on shared fresh water resources. Queen's Law J. pp. 342–369.

Burkhard, L.P., Armstrong, D.E., and Andren, A.W. 1985. Partitioning behaviour of polychlorinated biphenyls. Chemosphere, **14**: 1703–1716.

Caldwell, L.K. (*Editor*). 1988. Perspectives on ecosystem management for the Great Lakes: a reader. State University of New York Press, Albany, N.Y.

Carpenter, S.R., Jackson, L.J., Kitchell, J.F., and Stow, C.A. 1996. Organochlorine contaminants in the Great Lakes. Ecol. Appl. **6**: 971–974.

Christie, W.J. 1995. The ecosystem approach to managing the Great Lakes: the new ideas and problems associated with implementing them. Univ. Toledo Law Rev. **26**: 279–304.

Christie, W.J., Spangler, G.R., Loftus, K.H., Hartman, W.L., Colby, P.J., Ross, M.A., and Talhelm, D.R. 1987. A perspective on Great Lakes fish community rehabilitation. Can. J. Fish. Aquat. Sci. **44**(Suppl. 2): 486–499.

Colborn, T., and Clement, C. (*Editors*). 1992. Chemically-induced alterations in sexual and functional development: the wildlife–human connection. Princeton Scientific Publishing Co., Inc., Princeton, N.J.

Cooper, W.E. 1995. Risks of organochlorine contaminants to Great Lakes ecosystems are overstated. Ecol. Appl. **5**: 293–298.

Daly, H.B. 1993. Laboratory rat experiments show consumption of Lake Ontario salmon causes behavioral changes: support for wildlife and human research results. J. Great Lakes Res. **19**: 784–788.

Dar, E., Kanarek, M.S., Anderson, H.A., and Sonzogni, W.C. 1992. Fish consumption and reproductive outcomes in Green Bay, Wisconsin. Environ. Res. **59**: 189–201.

DeRosa, C., Johnson, B., and Gilbertson, M. 1995. Human health research to respond to the Great Lakes Water Quality Agreement. Great Lakes Res. Rev. **2**: 45–48.

Dunbar, M.J. 1980. The blunting of Occam's razor, or to hell with parsimony. Can. J. Zool. **58**: 123–128.

Egerton, F.N. 1985. Overfishing or pollution? Case history of a controversy on the Great Lakes. Great Lakes Fish. Comm. Tech. Rep. No. 41.

Eshenroder, R.L., Poe, T.P., and Olver, C.H. 1984. Strategies for Rehabilitation of Lake Trout in the Great Lakes: Proceedings of a

© 1997 NRC Canada

Conference on Lake Trout Research. Great Lakes Fish. Comm. Tech. Rep. No. 40.

Fein, G.G., Jacobson, J.L., Jacobson, S.W., Schwartz, P.M., and Dowler, J.K. 1984. Prenatal exposure to polychlorinated biphenyls: effects on birth size and gestational age. J. Pediatr. **105**: 315–320.

Fox, G.A. 1991. Practical causal inference for ecoepidemiologists. J. Toxicol. Environ. Health, **33**: 359–373.

Fox, G.A., Collins, B., Hayakawa, E., Weseloh, D.V., Ludwig, J.P., Kubiak, T.J., and Erdman, T.C. 1991. Reproductive outcomes in colonial fish-eating birds: a biomarker for developmental toxicants in Great Lakes food chains. II. Spatial variation in the occurrence and prevalence of bill defects in young double-crested cormorants in the Great Lakes, 1979–1987. J. Great Lakes Res. **17**: 158–167.

Francis, G.R., and Regier, H.A. 1995. Barriers and bridges to the restoration of the Great Lakes basin ecosystem. *In* Barriers and bridges for the renewal of ecosystems and institutions. *Edited by* L.H. Gunderson, C.S. Holling, and S.S. Light. Columbia University Press, New York. pp. 239–291.

Gilbertson, M. (*Editor*). 1996. Special section on cause–effect linkages. J. Great Lakes Res. **22**: 128–212.

Gilbertson, M., and Fox, G.A. 1977. Pollutant-associated embryonic mortality of Great Lakes herring gulls. Environ. Pollut. **12**: 211-216.

Gilbertson, M., and Schneider, R.S. (*Editors*). 1991. Special issue: International Joint Commission Workshop on Cause–Effect Linkages. J. Toxicol. Environ. Health, **33**: 359–639.

Gilbertson, M., and Schneider, R.S. (*Editors*). 1993. Special section on cause–effect linkages. J. Great Lakes Res. **19**: 720–807.

Gilbertson, M., Morris, R.D., and Hunter, R.A. 1976. Abnormal chicks and PCB residue levels in eggs of colonial birds on the lower Great Lakes (1971–1973). Auk, **93**: 434–442.

Gilbertson, M., Kubiak, T., Ludwig, J.P., and Fox, G.A. 1991. Great Lakes embryo mortality, edema, and deformities syndrome (GLEMEDS) in colonial fish-eating birds: similarity to chick-edema disease. J. Toxicol. Environ. Health, **33**: 455–520.

Great Lakes Water Quality Board. 1993. Legislative and regulatory considerations for virtual elimination of persistent toxic substances. International Joint Commission, Windsor, Ont.

Harris, G.E., Metcalfe, T.L., Metcalfe, C.D., and Huestis, S.Y. 1994. Embryotoxicity of extracts from Lake Ontario rainbow trout (*Oncorhynchus mykiss*) to Japanese medaka (*Oryzias latipes*). Environ. Toxicol. Chem. **13**: 1393–1403.

Holling, C.S. 1992. Cross-scale morphology, geometry, and dynamics of ecosystems. Ecol. Monogr. **62**: 447–502.

Holling, C.S., and Meffe, G.K. 1996. Command and control and the pathology of natural resource management. Conserv. Biol. **10**: 328–337.

Howdeshell, M.J., and Hites, R.A. 1996. Is the Hyde Park dump, near the Niagara River, still affecting the sediment of Lake Ontario? Environ. Sci. Technol. **30**: 969–974.

Humphrey, H.E.B. 1983. Population studies of PCBs in Michigan residents. *In* PCBs: human and environmental hazards. *Edited by* M. D'Itri and M.A. Kamrin. Butterworth, Boston, Mass. pp. 299–310.

International Joint Commission. 1969. Report to the International Joint Commission on the pollution of Lake Erie, Lake Ontario and the international section of the St. Lawrence River. Vol. 1. Summary. International Joint Commission, Windsor, Ont.

International Joint Commission. 1978. The ecosystem approach. Report of the Great Lakes Science Advisory Board to the International Joint Commission, Windsor, Ont.

International Law Association. 1967. Report of the Fifty-Second Conference held at Helsinki, August 14 to 20, 1966. International Law Association, London, U.K.

International Law Association. 1983. Report of the Sixtieth Conference held at Montreal, August 29 to September 4, 1982. International Law Association, London, U.K.

Jacobson, J. 1994. Children of Great Lakes fish consumers. *In* Applying Weight of Evidence: Issues and Practice; Proceedings of a Workshop of the International Joint Commission. *Edited by* M. Gilbertson and S. Cole-Misch. International Joint Commission, Windsor, Ont. pp. 9–15.

Jacobson, J.J., and Jacobson, S.W. 1996. Intellectual impairment in children exposed to polychlorinated biphenyls in utero. N. Engl. J. Med. **335**: 783–789.

Jones, P.D., Ankley, G.T., Best, D.A., Crawford, R., DeGalan, N., Giesy, J.P., Kubiak, T.J., Ludwig, J.P., Newsted, J.L., Tillitt, D.E., and Verbrugge, D.A. 1993. Biomagnification of bioassay derived 2,3,7,8-tetrachlorodibenzo-*p*-dioxin equivalents. Chemosphere, **26**: 1203–1212.

Kay, J.J., and Schneider, E. 1994. Embracing complexity: the challenge of the ecosystem approach. *In* Alternatives. Vol. 20. No. 3. Faculty of Environmental Studies, Waterloo, Ont. pp. 32–39.

Keith, J.A. 1966. Reproduction in a population of herring gulls (*Larus argentatus*) contaminated by DDT. J. Appl. Ecol. **3**(Suppl.): 57–70.

Kitchell, J.F. 1995. Toxic contaminants in the Great Lakes: a polythetic view of the issues and evidence. Ecol. Appl. **5**: 304–307.

Koonce, J.F. 1995. Challenges to ecosystem management in the Great Lakes. Forum, **1**: 4–5.

Kubiak, T.J., Harris, H.J., Smith, L.M., Schwartz, T.R., Stalling, D.L., Trick, J.A., Sileo, L., Docherty, D.E., and Erdman, T.C. 1989. Microcontaminants and reproductive impairment of the Forster's tern on Green Bay, Lake Michigan: 1983. Arch. Environ. Contam. Toxicol. **18**: 706–727.

Kuhn, T.S. 1962. The structure of scientific revolutions. University of Chicago Press, Chicago, Ill.

Leonardelli, S. 1995. Industrial releases within the Great Lakes basin: an evaluation of NPRI and TRI data. Environment Canada, Ontario Region, Toronto, Ont.

Loftus, K.H., and Regier, H.A. 1972. Introduction to the Proceedings of the 1971 Symposium on Salmonid Communities in Oligotrophic Lakes. J. Fish. Res. Board Can. **29**: 613–616.

Lonky, E., Reihman, J., Darvill, T., Mather, J., Sr., and Daly, H. 1996. Neonatal behavioral assessment scale performance in humans influenced by maternal consumption of environmentally contaminated Lake Ontario fish. J. Great Lakes Res. **22:** 198–212.

Ludwig, J.P. 1996. Contaminants effected widespread changes of Great Lakes populations and communities. Ecol. Appl. **6**: 962–965.

Ludwig, J.P., and Tomoff, C.S. 1966. Reproductive success and insecticide residues in Lake Michigan herring gulls. Jack-Pine Warbler, **44**: 77–84.

Ludwig, J.P., Giesy, J.P., Summer, C.L., Bowerman, W.W., Heaton, S., Aulerich, R., Bursian, S., Auman, H.J., Jones, P.D., Williams, L.L., Tillitt, D.E., and Gilbertson, M. 1993. A comparison of water quality criteria for the Great Lakes based on human and wildlife health. J. Great Lakes Res. **19**: 789–807.

Ludwig, J.P., Kurita-Matsuba, H., Auman, H.J., Ludwig, M.E., Summer, C.L., Giesy, J.P., Tillitt, D.E., and Jones, P.D. 1996. Deformities, PCBs and TCDD-equivalents in double-crested cormorants (*Phalacrocorax auritus*) and Caspian terns (*Hydroprogne caspia*) of the upper Great Lakes 1986–1991: testing a cause–effect hypothesis. J. Great Lakes Res. **22**: 172–197.

Mac, M.J., and Edsall, C.C. 1991. Environmental contaminants and the reproductive success of lake trout in the Great Lakes: an epidemiological approach. J. Toxicol. Environ. Health, **33**: 375–394.

© 1997 NRC Canada

National Research Council of the United States and the Royal Society of Canada. 1985. The Great Lakes Water Quality Agreement: an evolving instrument for ecosystem management. National Academy Press, Washington, D.C.

Paneth, N. 1991. Human reproduction after eating PCB-contaminated fish. Health Environ. Dig. **5**(8): 4–6.

Popper, K.R. 1962. Conjectures and refutations: the growth of scientific knowledge. Basic Books, Inc., New York.

Popper, K.R. 1968. The logic of scientific discovery. Harper and Row, New York.

Regier, H.A. 1979. Changes in species composition of Great Lakes fish communities caused by man. *In* Transactions of the 44th North American Wildlife and Natural Resources Conference, Toronto, Ont., March 24–28, 1979. *Edited by* K. Sabol. Wildlife Management Institute, Washington, D.C. pp. 558–566.

Regier, H.A., and Loftus, K.H. 1972. Effects of fisheries exploitation on salmonid communities in oligotrophic lakes. J. Fish. Res. Board Can. **29**: 959–968.

Silkworth, J.B., Cutler, D.S., Keefe, P.W.O., and Lipinskas, T. 1993. Potentiation and antagonism of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin effects in a complex mixture. Toxicol. Appl. Pharmacol. **119**: 236–247.

Simonich, S.L., and Hites, R.A. 1995. Global distribution of persistent organochlorine compounds. Science (Washington, D.C.), **269**: 1851–1854.

Smith, D.W. 1995. Are PCBs in the Great Lakes approaching a "new equilibrium"? Environ. Sci. Technol. **29**: 42A–46A.

Smith, S.H. 1972. Factors of ecological succession in oligotrophic fish communities of the Laurentian Great Lakes. J. Fish. Res. Board Can. **29**: 717–730.

Smith, I.R., Marchant, B., Van den Heuvel, M.R., Clemons, J.H., and Frimeth, J. 1994. Embryonic mortality, bioassay derived 2,3,7,8-tetrachlorodibenzo-*p*-dioxin equivalents, and organochlorine contaminants in Pacific salmon from Lake Ontario. J. Great Lakes Res. **20**: 497–509.

Smith, L.M., Schwartz, T.R., Feltz, K., and Kubiak, T.J. 1990. Determination and occurrence of AHH-active polychlorinated biphenyls, 2,3,7,8-tetrachlorodibenzo-*p*-dioxin and 2,3,7,8-tetrachlorodibenzofuran in Lake Michigan sediment and the question of their significance. Chemosphere, **21**: 1063–1085.

Spangler, G.R., Loftus, K.H., and Christie, W.J. 1987. Introduction to the International Symposium on Stock Assessment and Yield Prediction (ASPY). Can. J. Fish. Aquat. Sci. **44**(Suppl. 2): 7–9.

Sprunt, A., Robertson, W.B., Jr., Postupalsky, S., Hensel, R.J., Knoder, C.E., and Ligas, F.J. 1973. Comparative productivity of six bald eagle populations. Trans. N. Am. Wildl. Nat. Resour. Conf. **38**: 96–108.

Stow, C.A., Carpenter, S.R., Eby, L.A., Amrhein, J.F., and Hesselberg, R.J. 1995. Evidence that PCBs are approaching stable concentrations in Lake Michigan fishes. Ecol. Appl. **5**: 248–260.

Susser, M. 1986. The logic of Sir Karl Popper and the practice of epidemiology. Am. J. Epidemiol. **12**: 711–718.

Tillitt, D.E., Ankley, G.T., Giesy, J.P., Ludwig, J.P., Kurita Matsuba, H., Weseloh, D.V., Ross, P.S., Bishop, C.A., Sileo, L., Stromberg, K., Larson, J., and Kubiak, T.J. 1992. Polychlorinated biphenyl residues and egg mortality in double-crested cormorants from the Great Lakes. Environ. Toxicol. Chem. **11**: 1281–1288.

U.S. Environmental Protection Agency. 1995. Final water quality guidance for the Great Lakes system; final rule. Fed. Reg. **60**(56): 15366–15425. U.S. Environmental Protection Agency, Washington, D.C.

vom Saal, F.S., Nagel, S.C., Palanza, P., Boechler, M., Parmigiani, S., and Welshons, W.V. 1995. Estrogenic pesticides: binding relative to estradiol in MCF-7 cells and effects of exposure during fetal life on subsequent territorial behaviour in male mice. Toxicol. Lett. **77**: 343–350.

Yamashita, N., Tanabe, S., Ludwig, J.P., Kurita, H., Ludwig, M.E., and Tatsukawa, R. 1993. Embryonic abnormalities and organochlorine contamination in double-crested cormorants (*Phalacrocorax auritus*) and Caspian terns (*Hydroprogne caspia*) from the upper Great Lakes. Environ. Pollut. **79**: 163–173.

© 1997 NRC Canada

DISCUSSION

Comment — An improved link between industry, management and science: review of case history of the southwestern Gulf of St. Lawrence snow crab fishery[1,2]

Bernard Sainte-Marie

Introduction

A recent review by Loch et al. (1995) portrayed the snow crab (*Chionoecetes opilio*) exploitation in the southwestern Gulf of Saint Lawrence (eastern Canada) as an exemplary fishery. Industry involvement, better management practices, a unique method for evaluating exploitable biomass, and major insights into the biology of *C. opilio* were said to have contributed to rebuilding this snow crab stock after a 1987–1989 fishery collapse. I am compelled to reply to Loch and co-authors for two reasons. First, their review contains several unsubstantiated claims and is based on incomplete citation of literature. My commentary addresses only those scientific issues that are critical to resource management and conservation, drawing on information and literature available to the authors in 1994. Second, their review leaves the impression that directed human intervention was a major factor in the increase of the snow crab stock of the southwestern Gulf. While applauding recent advances in science and management, I point out that other snow crab fisheries in eastern Canada also declined between 1985 and 1989 and since then have fared just as well as the snow crab fishery in the southwestern Gulf. This may to a large extent reflect the effects of natural population fluctuations and of environmental change on the levels of fishery recruitment, rather than the effects of regional exploitation and management initiatives.

Received September 4, 1996. Accepted January 22, 1997.
J13644

B. Sainte-Marie. Division des invertébrés et de la biologie expérimentale, Institut Maurice-Lamontagne, ministère des Pêches et des Océans, 850, route de la Mer, C.P. 1000, Mont-Joli, QC G5H 3Z4, Canada. e-mail: b_stemarie@qc.dfo.ca

[1] Paper by J.S. Loch, M. Moriyasu, and J.B. Jones. 1995. Aquat. Living Resour. **8**: 253–265.

[2] Editor's note: The *Canadian Journal of Fisheries and Aquatic Sciences* does not normally consider correspondence upon papers that appear in other journals. However, the subject of this commentary was published in a journal that has no forum for discussion. The issues aired in this exchange appear to be of sufficient interest to resource biologists and managers that any barrier to potentially fruitful public debate seems inappropriate. Hence, *CJFAS* has provided the necessary forum in this case, and so informed *Aquatic Living Resources* of its intention.

The science

Terminal moult

Justifiably so, the concept of a terminal moult for male *C. opilio* is presented by Loch et al. (1995) as one of the single most important contributions to snow crab research in the past decade. After recognizing terminal moult as a fact (p. 256), the authors go on to state that "definitive evidence of a terminal moult remains controversial" (p. 259). There was indeed much debate and controversy over terminal moult up until 1991, largely for lack of supporting evidence (Dawe et al. 1991, and references therein). However, the idea that male *C. opilio* undergo a terminal moult is now accepted throughout eastern Canada and elsewhere, owing to the accumulation of convincing evidence from field (e.g., Comeau et al. 1991; Yamasaki and Kuwahara 1991; Sainte-Marie and Hazel 1992) and biochemical and histological (e.g., Bataller 1992; Cormier et al. 1992; Halcrow and Steel 1992) studies.

Loch et al. (1995) initially report that "it has become apparent that most males reach their terminal moult around 95 mm carapace width (CW)" (p. 256). This statement may suggest that a substantial fraction of males reach the legal size of 95 mm CW and that the range of body sizes for terminally moulted males is rather narrow. However, to date there still is no firm estimate of the proportion of males in a given year-class that cease to grow before they reach the legal size. Some field evidence indicates that at least 30–40% of males undergo terminal moult to sublegal size and, thus, never recruit to the fishery (Sainte-Marie and Dufour 1994). It is also possible that the proportion of males that reach legal size varies among year-classes, owing to density-dependent processes (Comeau et al. 1991). Further on in their review, Loch et al. (1995) state more specifically that "the size at which terminal moult occurs

© 1997 NRC Canada

varies from 50 to 120 mm CW" (p. 260). The meaning of this phrase is unclear: 50–120 mm CW either refers to the size range for males immediately prior to terminal moult, or it refers to the size range for males after terminal moult as given by Comeau and Conan (1992). The former interpretation would be consistent with Loch et al.'s Fig. 4 showing terminally moulted males of ≈58–137 mm CW. In any case, the range of body sizes given by Loch et al. (1995) is substantially less than what has been documented for terminally moulted males in Gulf populations of *C. opilio*, i.e., CWs of 40 mm (Sainte-Marie and Hazel 1992) to at least 150 mm (Bailey and Elner 1989; Sainte-Marie et al. 1992). Such extensive phenotypic or genotypic plasticity in size at terminal moult must be recognized, because it could have considerable implications for management (e.g., Bailey and Elner 1989), and elucidation of the underlying cause(s) represents one of the greatest challenges to snow crab science.

According to Loch et al. (1995), *C. opilio* survive "4 years or so" (p. 256), "about 5 years" (p. 260) or "3–4 years" (p. 261) after terminal moult. The authors also vaguely refer to changes in exoskeleton condition following terminal moult, which may affect market value of snow crab, and then state that ". . . the exploitable biomass can either be taken quickly in one or two years or extended over a larger period." (p. 261). However, there exists explicit information on temporal changes in exoskeleton condition and life expectancy for terminally moulted males, which allows inferences about the period of time that terminally moulted male snow crabs are available for exploitation. It is known that prerecruit males moult to legal size in the late winter or spring (e.g., Moriyasu et al. 1987; Conan et al. 1990; Sainte-Marie and Hazel 1992) and then require up to 5–6 months to fully harden, grow in flesh content (Watson 1971; O'Halloran and O'Dor 1988; Taylor et al. 1989), and disperse from moulting grounds (Bouchard et al. 1985). Consequently, males are not desirable or fully available to spring–summer fisheries until the year following their terminal moult. On the basis of a tag–recapture study conducted on a commercially unexploited population of *C. opilio* in the Saguenay Fjord, the life expectancy of males after terminal moult can reach up to 5–6 years (Sainte-Marie and Dufour 1994). However, a gradual deterioration in appearance and overall condition (hardiness, exoskeleton rigidity, number of missing limbs) causes males to become less catchable and to lose much of their prime market value within ≈4 years of their terminal moult. Thus, legal-sized males are fully available and valuable to spring–summer fisheries only for 3 years after their terminal moult (Sainte-Marie and Dufour 1994).

Reproduction

Because recent research indicates that female *C. opilio* in the Gulf of Saint Lawrence have a biennial instead of an annual reproductive cycle, and produce at most three egg clutches in their life-span, Loch et al. (1995) conclude that the "populations' reproductive potential is very low compared to what was believed and indicates the extreme fragility of the stock" (p. 260). In fact, these new findings only imply that biologists may have previously overestimated female lifetime fecundity. Since exploitation of female *C. opilio* is prohibited, the degree to which the southwestern Gulf snow crab stock is fragile really will depend on several other factors, for example, the maintenance of female reproductive potential in the face of moderate to high exploitation rates for large males (see below), larval production and survival rates relative to carrying capacity of benthic habitat for settlement and metamorphosis of larvae and growth of juveniles, the existence and importance of larval or juvenile subsidy from other stocks, and juvenile mortality rates. Given that information on these processes is scant or nonexistent, any statement on stock fragility at this time is pure speculation.

Recruitment and exploitation strategies

Loch et al. (1995) report for southwestern Gulf *C. opilio* "that recruitment of the 30 mm CW size group has occurred only twice (1988 and 1994) within the past 6 years of surveys" (p. 261) and then credit myself with having said that the same pattern was observed in the northern Gulf of Saint Lawrence. However, the 1993 stock status report for the northern Gulf (Sainte-Marie and Dufour 1994) tells a somewhat different story of recruitment variability for *C. opilio*. On the basis of a growth model and a time series of annual beam trawl surveys it was inferred that there was very little recruitment to northern Gulf populations (i.e., little settlement and (or) poor survival of early benthic instars) from 1977 to 1979 and again from 1985 to 1987. The 1977–1979 recruitment trough was undoubtedly responsible for the 1987–1989 fishery collapse, while the 1985–1987 recruitment trough was projected to affect fisheries starting in 1995. There was average to strong recruitment to populations from 1980 to 1984 and from 1988 to 1992, resulting in substantial recruitment to the 30 mm CW size group (i.e., post-settlement instar VII) from 1984 to 1988 and from 1992 to 1995. Troughs and waves of recruitment are seen in other populations of *C. opilio* in eastern Canada and may betray population cycles or fluctuations that are hypothetically either endogenous, generated by predators, or induced by the environment (Elner and Bailey 1986; Robichaud et al. 1989; Comeau et al. 1991; Sainte-Marie and Dufour 1994; Tremblay et al. 1994).

Detailed analyses of snow crab population dynamics during the early benthic stages are not possible for the southwestern Gulf stock, because the trawl that is used there for stock surveys does not adequately sample the smallest crabs (Anonymous 1994*a*). Nevertheless, the 1988 size frequency distribution for male snow crab in the southwestern Gulf (Fig. 4 in Loch et al. 1995) shows modes at ≈30, ≈40, ≈52, and ≈70 mm CW, representing four successive instars with annual intermoult periods, that clearly indicate that recruitment to the 30 mm CW size group occurred not only in 1988, but also in 1987, 1986, and 1985 as in the northern Gulf. It is the ≈70 mm CW size group seen in 1988, which, through two successive moults, first contributed to the 1991 upturn in catch rates and landings for the spring fishery in the southwestern Gulf. Thus, Loch et al. (1995) underestimated the contribution of recruitment to the increase in commercial biomass and, further, the southwestern Gulf fishery is not in its strict sense a "pulse fishery" (p. 263), because recruitment cannot be attributed to a single sporadic year-class.

With respect to interannual variability in fishery recruitment for *C. opilio*, Loch et al. (1995) state that "the level of current male parental stock biomass does not seem to affect or determine future recruitment instead, environmental factors seem to have a much larger role to play" (p. 261). This conclusion is not

© 1997 NRC Canada

supported by any of the data or references cited by Loch et al. (1995). The male parental stock could conceivably affect future fishery recruitment in either or both of two ways. One is through reproduction, an issue that Loch et al. (1995) indirectly address in a convoluted overview of the impact of carapace size limits and selective harvesting of large males on population reproductive potential, concluding as others had before (Elner and Beninger 1992) that "further research must be pursued to fully understand the mechanism of mating and reproductive potential of male crab" (p. 259). Nevertheless, there is some evidence that ovigerous female *C. opilio* are less fecund in exploited than in virgin populations (Taylor 1988) and that fecundity of large adult females in exploited populations is abnormally low (Elner and Gass 1984; Sainte-Marie 1993), owing possibly to sperm limitation for lack of sufficient numbers of large mates (Sainte-Marie 1993). Severe reduction of the male parental stock through exploitation could conceivably result in depressed female fecundity rates, which might eventually translate into reduced recruitment. The male parental stock might also modify the levels of fishery recruitment through any of a variety of hypothesized density-dependent mechanisms affecting suvivorship and growth of prerecruits, including competition for resources (Waiwood and Elner 1982; Tremblay et al. 1994), growth stunting (Conan et al. 1992), and cannibalism (Comeau and Conan 1992). Thus, although temporal heterogeneity of the environment may explain some measure of interannual variability in snow crab recruitment, as discussed above and below, it certainly is premature to discount the role of intrinsic or fishery-related fluctuations in the abundance of adult males for future stock condition.

Industry, management, science, and stock condition

The 1987–1989 collapse of the snow crab fishery in the southwestern Gulf of Saint Lawrence has widely been attributed to overfishing (e.g., Hare and Dunn 1993; Jamieson 1993), an interpretation that Loch et al. (1995) do not either explicitly support or dispel. Furthermore, Loch et al. (1995) argue that the stock subsequently recovered steadily owing to (*i*) a recruitment pulse of small crab in 1988, (*ii*) a better understanding of the biology of snow crab and the development of an accurate and reliable stock assessment methodology, (*iii*) the involvement of industry leaders, and (*iv*) close collaboration between industry and the Department of Fisheries and Oceans "to develop a management approach based upon scientific advice to climb out of the trough" (p. 253). However, this interpretation may underestimate the importance of natural events and overemphasize human influence on stock collapse and recovery.

In eastern Canada, there are also major snow crab fisheries in the northern Gulf of Saint Lawrence (6880 metric tons (t) in 1994) and around Newfoundland (28 990 t in 1994, D.M. Taylor, Science Branch, Department of Fisheries and Oceans, St. John's, Nfld., personal communication). Prior to 1990, the two areas experienced one period of rise and one period of decline in snow crab catch rates and landings, of similar magnitude to that recorded in the southwestern Gulf. Further, these events occurred almost synchronously in the northern and the southwestern Gulf (Sainte-Marie and Dufour 1994). In the Bonne Bay fjord, there also occurred a pronounced decline in the abundance of large male snow crabs from 1985 to 1989 (Ennis et al. 1990; Comeau et al. 1991), which closely paralleled change in the northern and southwestern Gulf. There is also evidence of a similar pattern in the abundance of large male snow crab in the Saguenay fjord (B. Sainte-Marie, unpublished data). However, neither fjord was commercially fished prior to 1994 and both have shallow external sills that limit or prevent exchange of bottom-dwelling snow crab with neighbouring exploited populations (Comeau et al. 1991; Sainte-Marie et al. 1992). Thus, evidence reviewed herein and in Taylor et al. (1994) suggests that declines in catches for fished populations of *C. opilio* may have resulted primarily from a natural decrease in the level of recruitment to legal size, not from overfishing per se.

From 1990 to 1994, overall, landings and (or) catch rates increased at least as rapidly in the northern Gulf, around Newfoundland, and off eastern Cape Breton as they did in the southwestern Gulf (compare data in Anonymous 1994*a*, 1994*b*; Sainte-Marie and Dufour 1994; Tremblay et al. 1994). This occurred under a variety of regional management regimes with different measures to control fishing effort. In particular, stock assessments for the northern Gulf, Newfoundland, and eastern Cape Breton areas were based mainly or exclusively on analysis of trends in commercial catch rates and effort. It is remarkable that major snow crab stocks in eastern Canada followed the same upward trend in spite of significant regional differences in scientific knowledge, stock assessment methodology, management policy, and (or) industry involvement and partnership with management and science. This suggests that pervasive natural change, more so than particular industry and management policies, was a major factor in the increase of exploitable snow crab biomass across eastern Canada.

For snow crab stocks in the northern and southwestern Gulf, the increase in exploitable biomass may have resulted primarily from recruitment to the fishery of 4 or 5 successive year-classes of moderate to strong importance (Sainte-Marie and Dufour 1994; see above). Moreover, fishery recruitment throughout eastern Canada was undoubtedly enhanced to some degree by an environmental context particularly favorable to *C. opilio*. From 1987 on, lower than average temperatures for the cold intermediate layer throughout the Gulf of Saint Lawrence caused a marked expansion of the surface area of soft bottoms bathed by waters <2–3°C (Swain 1993; Gilbert et al. 1996), which may represent prime snow crab habitat (Powles 1968; Brêthes et al. 1987). Expanding snow crab habitat could have resulted in reduced intraspecific competition and increased survival for fishery prerecruits and recruits. Concurrently, there occurred a drastic decline in predatory groundfish populations, including cod, *Gadus morhua* (Chouinard and Fréchet 1994), which is a known consumer of snow crab (Waiwood and Elner 1982; Robichaud et al. 1991). The demise of groundfish populations also resulted in the closure or down-scaling of many trawl and gill-net operations, potentially reducing incidental snow crab mortality. These alternative or complementary hypotheses for the surge in abundance and spatial distribution of snow crab throughout eastern Canada should be evaluated carefully.

Conclusion

It is true that in the southwestern Gulf, as elsewhere in eastern

© 1997 NRC Canada

Canada, industry has become increasingly aware of, and responsive to, the challenges of exploiting the fluctuating snow crab resource, management practices have been improved, and snow crab science has progressed in great leaps. The emerging ability to forecast snow crab abundance trends up to 5–10 years ahead is particularly laudable and holds promise for integrated long-term management schemes through partnership between the Department of Fisheries and Oceans and industry. However, despite the importance of advances in resource conservation institutions, there is strong evidence that the 1990–1994 increase in snow crab biomass in the southwestern Gulf resulted primarily from natural events. The review of Loch et al. (1995) does not offer a clear perspective on the relative roles of natural change and human intervention in the history and current condition of snow crab in the southwestern Gulf, or draw industry and management attention to outstanding scientific issues that must be addressed to further improve exploitation and management schemes. In particular, there is a pressing need for science to address several major questions regarding the following: the nature of fluctuations in snow crab recruitment; the pertinency, feasibility, and ways of dampening this variability or, otherwise, the implementation of optimal exploitation strategies for a fluctuating resource; the efficacy of current management practices in protecting population reproductive potential; and the medium- to long-term impact of selective exploitation of large males on population genetic diversity.

Acknowledgements

I thank M. Castonguay, R. Dufour, M. Fréchette, L. Gendron, H. Powles, L. Savard, and J.-M. Sévigny for discussion and comments on an earlier draft.

References

Anonymous. 1994*a*. General overview of Atlantic coast snow crab and report on the status of snow crab in the southern Gulf of St. Lawrence (fishing areas 12, 18, 19, 25 and 26). DFO Atlantic fisheries stock status report No. 94/1. Department of Fisheries and Oceans, Moncton, N.B.

Anonymous. 1994*b*. Newfoundland and Labrador snow crab stock status report. Newfoundland Science Branch, Department of Fisheries and Oceans, St. John's, Nfld.

Bailey, R.F.J., and Elner, R.W. 1989. Northwest Atlantic snow crab fisheries: lessons in research and management. *In* Marine invertebrate fisheries: their assessment and management. *Edited by* J.F. Caddy. John Wiley & Sons, New York. pp. 261–280.

Bataller, É. 1992. Étude histologique et ultrastructurale de l'organe Y chez les mâles immatures et matures du crabe des neiges (*Chionoecetes opilio*) (O. Fabricius) (Decapoda: Majidae). M.Sc. thesis, Université de Moncton, Moncton, N.B.

Bouchard, R., Brêthes, J.-C.F, and Desrosiers, G. 1985. Changes in size distribution of snow crabs (*Chionoecetes opilio*) in the southwestern Gulf of St. Lawrence. J. Northwest Atl. Fish. Sci. **7**: 67–75.

Brêthes, J.-C.F., Coulombe, F., Lafleur, P.-E., and Bouchard, R. 1987. Habitat and spatial distribution of early benthic stages of the snow crab *Chionoecetes opilio* O. Fabricius off the north shore of the Gulf of St. Lawrence. J. Crustacean Biol. **7**: 667–681.

Chouinard, G.A., and Fréchet, A. 1994. Fluctuations in cod stocks of the Gulf of St. Lawrence. ICES Mar. Sci. Symp. **198**: 121–139.

Comeau, M., and Conan, G.Y. 1992. Morphometry and gonad maturity of male snow crab, *Chionoecetes opilio*. Can. J. Fish. Aquat. Sci. **49**: 2460–2468.

Comeau, M., Conan, G.Y., Robichaud, G., and Jones, A. 1991. Life history patterns and population fluctuations of snow crab (*Chionoecetes opilio*) in the fjord of Bonne Bay on the west coast of Newfoundland, Canada, from 1983 to 1990. Can. Tech. Rep. Fish. Aquat. Sci. No. 1817.

Conan, G.Y., Elner, R.W., and Moriyasu, M. 1990. Review of the literature on life histories in the genus *Chionoecetes* in light of recent findings on growth and maturity in eastern Canada. *In* Proceedings of the International Symposium on King and Tanner crabs. Lowell Wakefield fisheries symposia series. Alaska Sea Grant report No. 90-04. Alaska Sea Grant College Program, Fairbanks, Alaska. pp. 163–179.

Conan, G.Y., Comeau, M., and Robichaud, G. 1992. Life history and fishery management of majid crabs: the case study of the Bonne Bay (Newfoundland) *Chionoecetes opilio* population. ICES C.M. 1992/K:21. International Council for the Exploration of the Sea, Copenhagen, Denmark.

Cormier, R.J., Fraser, A.R., Bailey, R.F.J., and Raymond, N. 1992. Hemolymph ecdysone concentration as a function of sexual maturity in the male snow crab (*Chionoecetes opilio*). Can. J. Fish. Aquat. Sci. **49**: 1619–1623.

Dawe, E.G., Taylor, D.M., Hoenig, J.M., Warren, W.G., Ennis, G.P., Hooper, R.G., Donaldson, W.E., Paul, A.J., and Paul, J.M. 1991. A critical look at the idea of terminal molt in male snow crabs (*Chionoecetes opilio*). Can. J. Fish. Aquat. Sci. **48**: 2266–2275.

Elner, R.W., and Bailey, R.F.J. 1986. Differential susceptibility of Atlantic snow crab, *Chionoecetes opilio*, stocks to management. *In* North Pacific Workshop on stock assessment and management of invertebrates. *Edited by* G.S. Jamieson and N. Bourne. Can. Spec. Publ. Fish. Aquat. Sci. No. 92. pp. 335–346.

Elner, R.W., and Beninger, P.G. 1992. The reproductive biology of snow crab, *Chionoecetes opilio*: a synthesis of recent contributions. Am. Zool. **32**: 524–533.

Elner, R.W., and Gass, C.A. 1984. Observations on the reproductive condition of female snow crabs from northwest Cape Breton Island, November 1983. CAFSAC research document No. 84/14. Canadian Atlantic Fisheries Scientific Advisory Committee, Dartmouth, N.S.

Ennis, G.P., Hooper, R.G., and Taylor, D.M. 1990. Changes in the composition of snow crab (*Chionoecetes opilio*) participating in the annual breeding migration in Bonne Bay, Newfoundland. Can. J. Fish. Aquat. Sci. **47**: 2242–2249.

Gilbert, D., Pettigrew, B., Swain, D., and Couture, M. 1996. State of the Gulf of St. Lawrence: oceanographic conditions in 1994. Can. Data Rep. Hydrogr. Ocean Sci. No. 143.

Halcrow, K., and Steel, C.G.H. 1992. Cuticular repair in the mature male snow crab, *Chionoecetes opilio* (Majidae: Crustacea): relation to ecdysteroids. Can. J. Zool. **70**: 314–319.

Hare, G., and Dunn, D.L. 1993. A retrospective analysis of the Gulf of St. Lawrence snow crab (*Chionoecetes opilio*) fishery 1965–1990. *In* Perspectives on Canadian marine fisheries management. *Edited by* L.S. Parsons and W.H. Lear. Can. Bull. Fish. Aquat. Sci. No. 226. pp. 177–192.

Jamieson, G.S. 1993. Marine invertebrate conservation: evaluation of fisheries over-exploitation concerns. Am. Zool. **33**: 551–567.

Loch, J.S., Moriyasu, M., and Jones, J.B. 1995. An improved link between industry, management and science: review of case history of the Southwestern Gulf of St. Lawrence snow crab fishery. Aquat. Living Resour. **8**: 253–265.

Moriyasu, M., Conan, G.Y., Mallet, P., Chiasson, Y., and Lacroix, H. 1987. Growth at molt, molting season and mating of snow crab (*Chionoecetes opilio*) in relation to functional and morphomet-

© 1997 NRC Canada

ric maturity. ICES C.M. 1987/K:21. International Council for the Exploration of the Sea, Copenhagen, Denmark.

O'Halloran, M.J., and O'Dor, R.K. 1988. Molt cycle of male snow crabs, *Chionoecetes opilio*, from observations of external features, setal changes, and feeding behavior. J. Crustacean Biol. **8**: 164–176.

Powles, H.W. 1968. Distribution and biology of the spider crab *Chionoecetes opilio* in the Magdalen Shallows, Gulf of Saint Lawrence. Manuscript report series No. 997. Fisheries Research Board of Canada, Ottawa, Ont.

Robichaud, D.A., Bailey, R.F.J., and Elner, R.W. 1989. Growth and distribution of snow crab, *Chionoecetes opilio*, in the southeastern Gulf of St. Lawrence. J. Shellfish Res. **8**: 13–23.

Robichaud, D.A., Elner, R.W., and Bailey, R.F.J. 1991. Differential selection of crab *Chionoecetes opilio* and *Hyas* sp. as prey of sympatric cod *Gadus morhua* and thorny skate *Raja radiata*. Fish. Bull. **89**: 669–680.

Sainte-Marie, B. 1993. Reproductive cycle and fecundity of primiparous and multiparous female snow crab, *Chionoecetes opilio*, in the northwest Gulf of Saint Lawrence. Can. J. Fish. Aquat. Sci. **50**: 2147–2156.

Sainte-Marie, B., and Dufour, R. 1994. Snow crab in the estuary and northern Gulf of St. Lawrence. *In* Status report on invertebrates 1993: crustaceans and molluscs on the Québec coast and northern shrimp in the Estuary and Gulf of St. Lawrence. *Edited by* L. Savard. Can. Manuscr. Rep. Fish. Aquat. Sci. No. 2257. pp. 2–20.

Sainte-Marie, B., and Hazel, F. 1992. Moulting and mating of snow crabs, *Chionoecetes opilio* (O. Fabricius), in shallow waters of the northwestern Gulf of Saint Lawrence. Can. J. Fish. Aquat. Sci. **49**: 1282–1293.

Sainte-Marie, B., Lapointe, V., Archambault, D., and Dufour, R. 1992. Distribution spatiale et structure de taille du crabe des neiges, *Chionoecetes opilio* (O. Fabricius), dans le fjord du Saguenay. Can. Manuscr. Rep. Fish. Aquat. Sci. No. 2156.

Swain, D.P. 1993. Variation in September near-bottom temperatures in the southern Gulf of St. Lawrence, 1971–1992. DFO Atlantic fisheries research document No. 93/48. Department of Fisheries and Oceans, Ottawa, Ont.

Taylor, D.M. 1988. A review of the literature pertaining to the reproductive biology of the genus *Chionoecetes*. *In* Proceedings of the International Workshop on Snow Crab Biology, Montréal, Quebec, December 8–10, 1987. *Edited by* G.S. Jamieson and W.D. McKone. Can. Manuscr. Rep. Fish. Aquat. Sci. No. 2005. pp. 67–74.

Taylor, D.M., Marshall, G.W., and O'Keefe, P.G. 1989. Shell hardening in snow crabs *Chionoecetes opilio* tagged in soft-shelled condition. N. Am. J. Fish. Manage. **9**: 504–508.

Taylor, D.M., O'Keefe, P.G., and Fitzpatrick, C. 1994. A snow crab, *Chionoecetes opilio* (Decapoda, Majidae), fishery collapse in Newfoundland. Fish. Bull. **92**: 412–419.

Tremblay, M.J., Eagles, M.D., and Elner, R.W. 1994. Catch, effort and population structure in the snow crab fishery off eastern Cape Breton, 1978–1993: a retrospective. Can. Tech. Rep. Fish. Aquat. Sci. No. 2021.

Waiwood, K.G., and Elner, R.W. 1982. Cod predation of snow crab (*Chionoecetes opilio*) in the Gulf of St. Lawrence. *In* Proceedings of the International Symposium on the Genus *Chionoecetes*, Lowell Wakefield symposium series. Alaska Sea Grant report No. 82–10. Alaska Sea Grant College Program, Fairbanks, Alaska. pp. 499–520.

Watson, J. 1971. Ecdysis of the snow crab, *Chionoecetes opilio*. Can. J. Zool. **49**: 1025–1027.

Yamasaki, A., and Kuwahara, A. 1991. The terminal molt of male snow crab in the Japan Sea. Bull. Jpn. Soc. Sci. Fish. **57**: 1839–1844. (Can. Transl. Fish. Aquat. Sci. No. 5559.)

© 1997 NRC Canada

Reply — An improved link between industry, management and science: review of case history of the southwestern Gulf of St. Lawrence snow crab fishery[1]

M. Moriyasu, J.S. Loch, and J.B. Jones

Introduction

Loch et al. (1995) examined the recovery of the southwestern Gulf of St. Lawrence snow crab fishery after its nearly collapsed situation in 1988–1989 by highlighting industry involvement, better management practices, and a new survey method. This review did not elucidate underlying biological events but rather emphasized the relationship established among science, management, and industry, which resulted in a unique partnership between the Department of Fisheries and Oceans and industry. Nevertheless, some readers have the impression that the recovery of the snow crab stock was because of the good fishery management and not underlying biological events (Sainte-Marie 1997). In addition, there are some unclear statements that may mislead the reader about the general consensus of Canadian snow crab biologists on snow crab stock dynamics and management approach. Therefore, this response deals with further clarification of Loch et al. (1995) by including the most up to date accounts of developments in snow crab biology and stock assessment in eastern Canada.

Terminal molt

The existence of terminal molt in male snow crab (Conan and Comeau 1986) is the most important assumption for stock assessment in the southern Gulf of St. Lawrence. Arguments on the existence of terminal molt in the genus *Chionoecetes* have been exchanged among North American scientists since 1988 (Conan et al. 1988; Donaldson and Johnson 1988; Dawe et al. 1991). Strong evidence of the existence of terminal molt in *Chionoecetes opilio* has been provided since then (Comeau et al. 1991; Sainte-Marie and Hazel 1992; Yamasaki and Kuwahara 1991; Bataller 1992; Cormier et al. 1992; Halcrow and Steel 1992). However, Paul and Paul (1995) reported the molting of functionally mature males of *Chionoecetes bairdi*. Further study is needed to assess the existence of terminal molt in other species of the genus *Chionoecetes*.

In terms of size at terminal molt, we agree that several statements in Loch et al. (1995) were ambiguous. It is clear that males reach their terminal molt at a fairly wide size range. In some cases, at least 30–40% of males undergo terminal molt at a sublegal size (Sainte-Marie and Dufour 1994, 1995). We agree that the range (at which terminal molt occurred) shown in Fig. 4 is less than what has been reported for other areas in eastern Canada. Although the nature of variability in size at terminal molt is not yet known, we agree that such variability has significant implications for stock management.

Reproduction

Although phrases like "extreme fragility of the stock" are a value judgement, we believe that the current regulation of not exploiting females should be maintained until the reproductive and recruitment mechanisms and stock relationship are better understood.

Recruitment and exploitation strategies

Sainte-Marie and Dufour (1994) and Sainte-Marie et al. (1995) pointed out that recruitment observed in the Gulf of St. Lawrence was not comprised of a single size group but several size groups. Loch et al. (1995) stated only that two occurrences of 30-mm size groups were observed over 7 years, but did not further refer to the composition of these recruits. The most important message in Loch et al. (1995) is that the recruitment does not occur every year.

Southern Gulf snow crab can be described as a stock whose recent recruitment patterns are dominated by one or two very large year-classes that are evidenced in the sequence of recruitment data (Rothschild 1986). The snow crab fishery is a pulse fishery because catch rates show a transient increase over a relatively short period of time (5–6 years). The term pulserecruitment was also used by Daan et al. (1994) for the North Sea cod stock whose recruitment occurred over an even longer period. The term pulse fishery should not be interpreted as a single or sporadic year-class fishery.

There are no convincing published results available on the longevity of terminal molt males. Whether the longevity of males after terminal molt varies between 3 and 4 years or between 5 and 6 years does not change our general view. The

Received January 21, 1997. Accepted January 22, 1997.
J13837

M. Moriyasu,[2] J.S. Loch, and J.B. Jones. Department of Fisheries and Oceans, Science Branch, Maritime Region, P.O. Box 5030 Moncton, NB E1C 9B6, Canada.

[1] Comment by B. Sainte-Marie. 1997. Can. J. Fish. Aquat. Sci. **54**: 000–000.

[2] Author to whom all correspondence should be addressed. e-mail: moriyasum@gfc.dfo.ca

© 1997 NRC Canada

main point is to provide a choice to industry and fisheries managers whether fishers should exploit animals immediately after the recruitment to the fishery or expand the exploitation over several years.

Industry, management, science, and stock condition

The reasons for the 1987–1989 collapse are not the main issue of our paper. Our untested hypothesis was that the lack of recruitment and a very high exploitation rate of both hard- and soft-shelled males greatly decreased the biomass of exploitable terminal molt crabs. This high fishing mortality was accelerated by increased fishing effort (Chiasson et al. 1992). Sainte-Marie (1997) stated that "[t]his suggests that pervasive natural change, more so than particular regional industry and management policies, was a major factor in the increase of exploitable snow crab biomass across eastern Canada." We agree that practicing a very conservative management strategy was not a major factor in the fluctuation of exploitable crab biomass. However, conservative stock management strategies provide a tool to enhance the recruitment to the fishery by protecting white crabs when strong recruitment waves are upcoming, and to expand the harvesting duration by setting lower exploitation rates when the recruitment to the fishery becomes scarce. The cyclic nature of natural fluctuation in snow crab abundance cannot be controlled, but its magnitude can be controlled to a certain extent by setting sound management strategies.

We agree that natural fluctuations in recruitment cannot be ignored. We mentioned in our paper that the most important reason for this stock recovery was new recruitment: "The current high biomass level is exclusively due to the arrival of a good recruitment of juvenile crabs (about 30 mm)." As we thought that the collapse of the stock was due to the lack of recruitment, we attributed the recovery of the stock to the strong recruitment pulse together with an extremely conservative management approach taken by management and industry. We agree that these hypotheses should be assessed to understand how each factor contributed to the recovery of the stock.

We reiterate that the main purpose of our paper was to focus on the new relationship between industry and government, rather than to scrutinize scientifically the stock dynamics and underlying biological factors.

Acknowledgments

We thank Dr. E.M.P. Chadwick and Mr. M. Comeau (Gulf Fisheries Centre, Department of Fisheries and Oceans, Moncton, N.B.) for their critical review and valuable comments on the manuscript.

References

Bataller, E. 1992. Étude histologique et ultrastructural de l'organe Y chez les mâles immatures et matures du crabe des neiges (*Chionoecetes opilio*), (O. Fabricius) (Decapoda: Majidae). M.Sc. thesis, Université de Moncton, Moncton, N.B.

Chiasson, Y.J., Hébert, M., and Moriyasu, M. 1992. A retrospective look at the development and expansion of southwestern Gulf of St. Lawrence snow crab, *Chionoecetes opilio*, fishery. Can. Tech. Rep. Fish. Aquat. Sci. No. 1847.

Comeau, M., Conan, G.Y., Robichaud, G., and Jones, A. 1991. Life patterns and population fluctuations of snow crab (*Chionoecetes opilio*) in the fjord of Bonne Bay on the west coast of Newfoundland, Canada, from 1983 to 1990. Can. Tech. Rep. Fish. Aquat. Sci. No. 1817.

Conan, G.Y., and Comeau, M. 1986. Functional maturity of male snow crab (*Chionoecetes opilio*). Can. J. Fish. Aquat. Sci. **43**: 1710–1719.

Conan, G.Y., Comeau, M., Moriyasu, M., and Cormier, R. 1988. Reply to Donaldson and Johnson. Can. J. Fish. Aquat. Sci. **45**: 1501–1503.

Cormier, R.J., Fraser, A.R., Bailey, R.F.J., and Raymond, N. 1992. Hemolymph ecdysone concentration as a function of sexual maturity in the male snow crab (*Chionoecetes opilio*). Can. J. Fish. Aquat. Sci. **49**: 1619–1623.

Daan, N., Heessen, H.J.L., and Pope, J.G. 1994. Changes in the North Sea cod stock during the twentieth century. ICES Mar. Sci. Symp. **198**: 229–243.

Dawe, E.G., Taylor, D.M., Hoenig, J.H., Warren, W.G., Ennis, G.P., Hooper, R.G., Donaldson, W.E., Paul, A.J., and Paul, J.M. 1991. A critical look at the idea of terminal molt in male snow crab (*Chionoecetes opilio*). Can. J. Fish. Aquat. Sci. **48**: 2266–2275.

Donaldson, W.E., and Johnson, B.A. 1988. Some remarks on "Functional maturity and terminal molt of male snow crab, *Chionoecetes opilio*" by Conan and Comeau. Can. J. Fish. Aquat. Sci. **45**: 1499–1501.

Halcrow, K., and Steel, C.G.H. 1992. Cuticular repair in the mature male snow crab, *Chionoecetes opilio* (Majidae: Crustacea): relation to ecdysteroids. Can. J. Zool. **70**: 314–319.

Loch, J.S., Moriyasu, M., and Jones, J.B. 1995. An improved link between industry, management and science: review of case history of the southwestern Gulf of St. Lawrence snow crab fishery. Aquat. Living Resour. **8**: 253–265.

Paul, A.J., and Paul, J.M. 1995. Molting of functionally mature male *Chionoecetes bairdi* Rathbun (Decapoda: Majidae) and changes in carapace and chela measurements. J. Crustacean Biol. **15**: 686–692.

Rothschild, B.J. 1986. Dynamics of marine fish populations. Harvard University Press, Cambridge, U.K.

Sainte-Marie, B. 1997. Comment — An improved link between industry, management and science: review of case history of the southwestern Gulf of St. Lawrence snow crab fishery. Can. J. Fish. Aquat. Sci. **54**: 496–500.

Sainte-Marie, B., and Dufour, R. 1994. Crabe des neiges de l'estuaire et du nord du golfe Saint-Laurent (zones de pêche 13, 14, 15, 16 et 17). *In* Rapport sur l'état des invertébrés en 1993 : crustacés et mollusques des côtes du Québec et crevette nordique de l'estuaire et du golfe du Saint-Laurent. *Edited by* L. Savard. Can. Manuscr. Rep. Fish. Aquat. Sci. No. 2257. pp. 2–23.

Sainte-Marie, B., and Dufour, R. 1995. Crabe des neiges de l'estuaire et du nord du golfe Saint- Laurent (zones de pêche 13, 14, 15, 16 et 17). *In* Rapport sur l'état des invertébrés en 1994 : crustacés et mollusques des côtes du Québec et crevette nordique et zooplancton de l'estuaire et du golfe du Saint-Laurent. *Edited by* L. Savard. Can. Manuscr. Rep. Fish. Aquat. Sci. No. 2323. pp. 2–27.

Sainte-Marie, B., and Hazel, F. 1992. Moulting and mating of snow crabs, *Chionoecetes opilio* (O. Fabricius), in shallow waters of the northwestern Gulf of Saint Lawrence. Can. J. Fish. Aquat. Sci. **49**: 1282–1293.

Sainte-Marie, B., Raymond, S., and Brêthes, J.-C. 1995. Growth and maturation of the benthic stages of male snow crab, *Chionoecetes opilio* (*Brachyura: Majidae*). Can. J. Fish. Aquat. Sci. **52**: 903–924.

Yamasaki, A., and Kuwahara, A. 1991. The terminal molt of male snow crab in the Japan Sea. Nippon Suisan Gakkaishi, **57**: 1839–1844.

© 1997 NRC Canada

ASSIGNMENT OF COPYRIGHT

Ms. No._____

To: NATIONAL RESEARCH COUNCIL OF CANADA ("NRC") Research Journals

From: _____

_____

_____

_____

(name and address of author signing this form)

This refers to the manuscript entitled: _____

_____

written by myself and the following additional author(s): _____

_____

for publication in: _____

1. I declare that the list of authors fairly conforms to academic conventions about who should be listed. I declare that the manuscript, or its equivalent, has not been submitted for publication elsewhere. If it is accepted for publication by the National Research Council of Canada, I will ensure that it, or its equivalent, is not submitted for publication elsewhere.

2. I declare that I am (check ONE):

☐ (a) the sole owner of the copyright relating to the manuscript,	and →	I **assign** to NRC the entire copyright.
☐ (b) a part owner of the copyright, in conjunction with interests held by co-authors or their employers,	and →	I **assign** to NRC my entire interest in the copyright, and I expect the co-authors to also sign a form like this one.
☐ (c) an agent of my employer with authority to assign the copyright owned by the employer, who is: _____,	and →	I **assign** to NRC, on behalf of my employer, my employer's entire interest in the copyright relating to the manuscript, and I expect the co-authors to also sign a form like this one.
☐ (d) a public servant in Canada (federal, provincial or territorial) or the United Kingdom, so that the copyright is owned by Her Majesty,	and →	I have obtained the approval of my employer, according to the terms of my employment, to grant NRC **non-exclusive permission** to publish the manuscript and administer the copyright.
☐ (e) a public servant of the United States of America, and subject to the law that creates no U.S. copyright for the manuscript,	and →	I **assign** to NRC, on behalf of my employer, my employer's entire interest in any copyright that may exist outside the U.S., and I expect the co-authors to sign a form like this one.

3. I understand that my submission of this document does not guarantee publication, and that publication will not occur until **all authors** have provided a copyright transfer. If the manuscript is not published within 2 years after I sign this, the assignment is revoked.

Signed at _____ on _____

(City and Province/State) (Date)

_____

(Signature)

[*Standard NRC form; alterations not acceptable.*]

Comments on Copyright Transfer
(with extracts from the *Copyright Act*, Revised Statutes of Canada 1985, chapter C-42, as amended)

The publisher is not permitted to publish the manuscript without permission or an assignment of ownership of copyright. For greatest flexibility, and to be prepared to deal with future uses of the published material, the publisher requires ownership.

The owner may assign the right, but that assignment must be in writing:

13.(4) The owner of the copyright in any work may assign the right, either wholly or partially, and either generally or subject to territorial limitations, and either for the whole term of the copyright or for any other part thereof, and may grant any interest in the right by licence, but no assignment or grant is valid unless it is in writing signed by the owner of the right in respect of which the assignment or grant is made, or by his duly authorized agent.

The owner may be the author, but in many cases, the owner is the author's employer:

13.(3) Where the author of a work was in the employment of some other person under a contract of service or apprenticeship and the work was made in the course of his employment by that person, the person by whom the author was employed shall, in the absence of any agreement to the contrary, be the first owner of the copyright, but where the work is an article or other contribution to a newspaper, magazine or similar periodical, there shall, in the absence of any agreement to the contrary, be deemed to be reserved to the author a right to restrain the publication of the work, otherwise than as part of a newspaper, magazine or similar periodical.

The only exception to taking ownership arises when the manuscript is covered by Crown copyright owned by a Canadian government.

12. Without prejudice to any rights or privileges of the Crown, where any work is, or has been, prepared or published by or under the direction or control of Her Majesty or any government department, the copyright in the work shall, subject to any agreement with the author, belong to Her Majesty and in that case shall continue for the remainder of the calendar year of the first publication of the work and for a period of fifty years following the end of that calendar year.

Governments, and in particular the federal government, do not have a simple mechanism for assigning ownership of this particular type of Crown asset. Therefore, when an author is a public servant so that there is Crown copyright in the manuscript, a simple permission to use is acceptable to the publisher, if it is granted by a person with appropriate authority.

CESSION DU DROIT D'AUTEUR

Document n° _____

Au : CONSEIL NATIONAL DE RECHERCHES DU CANADA («CNRC») - Presses scientifiques

De : _____

_____

_____

_____

(nom et adresse de l'auteur qui signe le présent formulaire)

La présente cession porte sur le document intitulé : _____

_____

rédigé par moi, en collaboration avec l'auteur/les auteurs suivant(s) : _____

_____

aux fins de publication dans : _____

1. Je déclare que la liste des auteurs ci-dessus respecte les pratiques usuelles de rédaction savante quant aux personnes dont le nom doit être cité. Je déclare que le document ou son équivalent n'a pas été soumis à quelque autre organisme aux fins de publication. Si le CNRC accepte de publier le document, je m'assurerai que celui-ci ou son équivalent n'est soumis à aucun autre organisme aux fins de publication sans en obtenir la permission préalable du CNRC.

2. Je déclare être (UN SEUL choix) :

☐	(a) le seul titulaire de droit d'auteur afférent au document,	et →	je **cède** le droit d'auteur intégral au CNRC.
☐	(b) un titulaire partite du droit d'auteur, de concert avec des coauteurs ou leurs employeurs,	et →	je **cède** ma part du droit d'auteur au CNRC et m'attends à ce que les coauteurs signent une entente comparable.
☐	(c) un agent de mon employeur ayant l'autorité de céder le droit d'auteur appartenant à l'employeur, nommément : _____,	et →	je **cède** au CNRC, au nom de mon employeur, la part entière du droit d'auteur afférent au document appartenant à mon employeur, et m'attends à ce que les coauteurs signent une entente comparable.
☐	(d) un fonctionnaire du Canada (fédéral, provincial ou territorial) ou du Royaume-Uni, donc le droit d'auteur appartient à Sa Majesté,	et →	j'ai obtenu de mon employeur, en vertu de mes conditions d'emploi, l'autorisation d'accorder au CNRC la **permission non exclusive** de publier le document et de gérer le droit d'auteur.
☐	(e) un fonctionnaire des États-Unis d'Amérique, donc sujet à la loi qui n'associe aucun droit d'auteur américain au document,	et →	je **cède** au CNRC, au nom de mon employeur, la part entière de quelque droit d'auteur en vigueur ailleurs qu'aux É.-U. appartenant à mon employeur, et m'attends à ce que les coauteurs signent une entente comparable.

3. Je comprends que la soumission du document ne garantit aucunement sa publication et que la publication ne pourra être réalisée tant que **tous les auteurs** n'auront pas signé une cession du droit d'auteur. Si le document n'est pas publié dans les deux années suivant ma signature des présentes, la cession sera automatiquement révoquée.

Signé à _____ (ville et province/état), le _____ (date)

_____

(signature)

[Formulaire normalisé du CNRC; aucune modification permise]

Commentaires sur la cession du droit d'auteur
(accompagnés d'extraits de la *Loi sur le droit d'auteur*, Lois révisées du Canada 1985, chapitre C-42 modifié)

L'éditeur ne peut publier un document sans la permission ou une cession du droit d'auteur. Pour des raisons de souplesse et en vue de l'utilisation future du document publié, l'éditeur doit devenir titulaire du droit d'auteur.

Le titulaire peut céder le droit, mais cette cession doit être faite par écrit :

13.(4) Le titulaire du droit d'auteur sur une oeuvre peut céder ce droit, en totalité ou en partie, d'une façon générale, ou avec des restrictions territoriales, pour la durée complète ou partielle de la protection; il peut également concéder, par une licence, un intérêt quelconque dans ce droit; mais la cession ou la concession n'est valable que si elle est rédigée par écrit et signée par le titulaire du droit qui en fait l'objet, ou par son agent dûment autorisé.

Le titulaire peut être l'auteur, mais dans plusieurs cas, le titulaire est l'employeur de l'auteur:

13.(3) Lorsque l'auteur est employé par une autre personne en vertu d'un contrat de louage de service ou d'apprentissage, et que l'oeuvre est exécutée dans l'exercice de cet emploi, l'employeur est, à moins de stipulation contraire, le premier titulaire du droit d'auteur; mais lorsque l'oeuvre est un article ou un autre contribution, à un journal, à une revue ou à un périodique du même genre, l'auteur, en l'absence de convention contraire, est réputé posséder le droit d'interdire la publication de cette oeuvre ailleurs que dans un journal, une revue ou un périodique semblable.

La seule exception dans la cession du droit d'auteur est lorsque le document est associé au droit d'auteur de la Couronne, qui appartient au gouvernement du Canada .

12. Sous réserve de tous les droits ou privilèges de la Couronne, le droit d'auteur sur les oeuvres préparées ou publiées par l'entremise, sous la direction ou la surveillance de Sa Majesté ou d'un ministère du gouvernement, appartient, sauf stipulation conclue avec l'auteur, à Sa Majesté et, dans ce cas, il subsiste jusqu'à la fin de la cinquantième année suivant celle de la première publication de l'oeuvre.

Les gouvernements, en particulier le gouvernement fédéral, ne disposent pas d'un mécanisme simple de cession de droit d'auteur pour ce genre de biens de la Couronne. C'est pourquoi lorsqu'un auteur est un fonctionnaire et que, par conséquent, le droit d'auteur appartient à la Couronne, une simple permission d'usage est suffisante pour l'éditeur, pour autant que celle-ci ait été octroyée par une personne jouissant de l'autorité nécessaire pour ce faire.

ANNOUNCEMENTS / ANNONCES

Numerical Methods in
Atmospheric and Oceanic Modelling

The André J. Robert Memorial Volume

Edited by/Rédaction : Charles Lin (McGill University), René Laprise (Université du Québec à Montréal), and/et Harold Ritchie (Atmospheric Environment Service, Environment Canada).

(approx./environ 624 pages, hardbound/, ISBN 0-9698414-4-2)
Canada $79.95 (CAN); Other countries/Autres pays $79.95 (US/É.-U.)

The Canadian Meteorological and Oceanographic Society (CMOS) in association with NRC Research Press, Canada Institute for Scientific and Technical Information, is pleased to announce the publication in April 1997 of a companion volume to Atmosphere-Ocean dedicated to the memory of Dr. André J. Robert, who passed away November 19, 1993. Dr. Robert had a long and distinguished career in numerical methods for atmospheric and fluid models. Among his major contributions are the first integration of a global spectral primitive equation model, the development of an efficient time filter, and pioneering studies in the use of the semi-implicit and semi-Lagrangian methods. The volume is a collection of refereed papers by leading scientists in the field of numerical modelling based on invited presentations.

Of the 27 papers, 25 are research papers reporting or reviewing recent original research findings. The other two papers present a historical perspective of numerical weather prediction and Dr. Robert's pioneering contributions to numerical modelling. The book also includes a supplement of four previously unpublished manuscripts by Dr. Robert.

This book will provide an up-to date comprehensive and long-lasting reference for students and researchers in numerical weather prediction, climate simulation, dynamic meteorology, and ocean modelling.

Please send your order to:

NRC Research Press
Building M-55
Ottawa, Ontario, Canada K1A 0R6
Telephone: (613) 993-0151
Fax: (613) 952-7656
E-mail: research.journals@nrc.ca

C'est avec plaisir que la Société canadienne de météorologie et d'océanographie (SCMO), en association avec les Presses scientifiques du CNRC, Institut canadien de l'information scientifique et technique, annonce la publication en avril 1997 d'un volume accompagnant Atmosphère-Océan et dédié à la mémoire du Dr André J. Robert, décédé le 19 novembre 1993. Le Dr Robert a eu une carrière longue et remarquable de recherche en méthodes numériques appliquées aux modèles des fluides et de l'atmosphère. Parmi ses contributions majeures, on retrouve la première intégration d'un modèle spectral global utilisant les équations primitives, le développement d'un filtre temporel efficace ainsi que des études fondamentales sur l'usage des méthodes semi-implicite et semi-lagrangienne. Le volume est une collection d'articles révisés par des comités de lecture, basés sur des conférences présentées par des scientifiques réputés.

Des 27 articles, 25 sont des articles de recherche rapportant ou faisant revue de découvertes originales récentes. Les deux autres articles présentent une perspective historique de la prévision numérique et des contributions originales du Dr Robert à la modélisation numérique. Le volume incluera aussi en supplément quatre manuscripts du Dr Robert, jusqu'à maintenant inédits.

Ce volume servira comme référence extensive et à jour pour les étudiants et les chercheurs en prévision numérique du temps, simulation climatique, météorologie dynamique et modèles des océans.

Prière d'envoyer votre commande à :

Les Presses scientifiques du CNRC
Édifice M-55
Ottawa (Ontario) Canada K1A 0R6
Téléphone : (613) 993-0151
Télécopieur : (613) 952-7656
Courrrier éléctronique : research.journals@nrc.ca

Le programme de Monographies du CNRC

Les presses scientifiques du CNRC de l'ICIST, au Conseil national de recherches du Canada, est un éditeur à but non lucratif qui publie des monographies et des revues en sciences de la vie, en sciences physiques et en recherche, en français ou en anglais. Les soumissions au programme de Monographies sont examinées par des spécialistes du domaine et révisées à la lumière de leurs commentaires. Seuls les documents approuvés par les examinateurs seront publiés. Les soumissions approuvées sont sujettes à des modifications, autant au point de vue du contenu que de la présentation.

Comité consultatif

R.H. Haynes, OC, MSRC, *Directeur scientifique, Le programme de Monographies du CNRC*, (Université York)
W.G.E. Caldwell, MRSC (Université de Western Ontario)
P.B. Cavers, (Université de Western Ontario)
G. Herzberg, CAC, MRS, MRSC (Conseil national de recherches du Canada)
K.U. Ingold, OC, MRS, MRSC (Conseil national de recherches du Canada)
W. Kaufmann (Rédacteur en chef émérite, Annual Reviews Inc.)
M. Lecours (Université Laval)
L.P. Milligan, MRSC (Université de Guelph)
G.G.E. Scudder, MRSC (The University of British Columbia)
E.W. Taylor, MRS (The University of Chicago)
B.P. Dancik, *Directeur général, Revues scientifiques du CNRC,* (Université d'Alberta)

Les soumissions au programme

Les propositions de documents aux fins de publication dans la collection de monographies du CNRC doivent être adressées à :

G.J. Neville
Conseil national de recherches du Canada
Les presses scientifiques du CNRC, Édifice M-55
Ottawa (Ontario) Canada
K1A 0R6

Téléphone : (613) 993-1513
Télécopieur : (613) 952-7656
Courrier électronique : gerry.neville@nrc.ca

NRC Monograph Publishing Program

The NRC Research Press, CISTI, of the National Research Council of Canada is a not-for-profit group that publishes monographs and journals in various branches of life sciences, physical sciences, and engineering in English or French. Publishing proposals and manuscripts submitted to the Monograph Program are peer-reviewed by specialists in the field and subject to revision in light of the reviewers' comments. Only manuscripts that have received positive peer review may be considered for publication. Contributions accepted for publication in this series are closely edited for both content and presentation.

Monograph Editorial Board

R.H. Haynes, OC, FRSC, *Editor, NRC Monograph Publishing Program* (York University)
W.G.E. Caldwell, FRSC (University of Western Ontario)
P.B. Cavers (University of Western Ontario)
G. Herzberg, CC, FRS, FRSC (National Research Council of Canada)
K.U. Ingold, OC, FRS (National Research Council of Canada)
W. Kaufmann (Editor-in-Chief Emeritus, Annual Reviews Inc.)
M. Lecours (Université Laval)
L.P. Milligan, FRSC (University of Guelph)
G.G.E. Scudder, FRSC (The University of British Columbia)
E.W. Taylor, FRS (The University of Chicago)
B.P. Dancik, *Editor-in-Chief, NRC Research Journals & Monographs* (University of Alberta)

Publication Proposals

Proposals for the NRC Monograph Publishing Program Series should be sent to

Gerald J. Neville
National Research Council of Canada
NRC Research Press, Building M-55
Ottawa, Ontario, Canada
K1A 0R6

Telephone: (613) 993-1513
FAX: (613) 952-7656
E-mail: gerry.neville@nrc.ca

PERSPECTIVES		PERSPECTIVES
R. Quentin Grafton and Harry W. Nelson	**474–482**	Fishers' individual salmon harvesting rights: an option for Canada's Pacific fisheries
Michael Gilbertson	**483–495**	Are causes knowable? Some consequences of successional versus toxicological interpretations of the Great Lakes Water Quality Agreement
DISCUSSION		DISCUSSION
Bernard Sainte-Marie	**496–500**	Comment — An improved link between industry, management and science: review of case history of the southwestern Gulf of St. Lawrence snow crab fishery
M. Moriyasu, J.S. Loch, and J.B. Jones	**501–502**	Reply — An improved link between industry, management and science: review of case history of the southwestern Gulf of St. Lawrence snow crab fishery

Reinhard Pienitz, John P. Smol, and David R.S. Lean	**330–346**	Physical and chemical limnology of 59 lakes located between the southern Yukon and the Tuktoyaktuk Peninsula, Northwest Territories (Canada)
Reinhard Pienitz, John P. Smol, and David R.S. Lean	**347–358**	Physical and chemical limnology of 24 lakes located between Yellowknife and Contwoyto Lake, Northwest Territories (Canada)
B.J. Shuter and K.K. Ing	**359–377**	Factors affecting the production of zooplankton in lakes
John D. Wehr and James H. Thorp	**378–395**	Effects of navigation dams, tributaries, and littoral zones on phytoplankton communities in the Ohio River
G.C. Mair, J.S. Abucay, D.O.F. Skibinski, T.A. Abella, and J.A. Beardmore	**396–404**	Genetic manipulation of sex ratio for the large-scale production of all-male tilapia, *Oreochromis niloticus*
Benjamin H. Letcher, James A. Rice, Larry B. Crowder, and Fred P. Binkowski	**405–414**	Size- and species-dependent variability in consumption and growth rates of larvae and juveniles of three freshwater fishes
David W. Au and Susan E. Smith	**415–420**	A demographic method with population density compensation for estimating productivity and yield per recruit of the leopard shark (*Triakis semifasciata)*
James S. Baxter, Eric. B. Taylor, Robert H. Devlin, John Hagen, and J. Donald McPhail	**421–429**	Evidence for natural hybridization between Dolly Varden (*Salvelinus malma*) and bull trout (*Salvelinus confluentus*) in a northcentral British Columbia watershed
Jörgen I. Johnsson, John Blackburn, W. Craig Clarke, and Ruth E. Withler	**430–433**	Does presmolt growth rate in steelhead trout (*Oncorhynchus mykiss*) and coho salmon (*Oncorhynchus kisutch*) predict growth rate in seawater?
Je-Chul Park, M. Aizaki, T. Fukushima, and A. Otsuki	**434–443**	Production of labile and refractory dissolved organic carbon by zooplankton excretion: an experimental study using large outdoor continuous flow-through ponds
Jeffrey T. Silverstein, Hisako Shimma, and Hiroshi Ogata	**444–451**	Early maturity in amago salmon (*Oncorhynchus masu ishikawai*): an association with energy storage
Mark J. Butler IV and William F. Herrnkind	**452–463**	A test of recruitment limitation and the potential for artificial enhancement of spiny lobster (*Panulirus argus*) populations in Florida
C.A. Stow, S.R. Carpenter, and R.C. Lathrop	**464–473**	A Bayesian observation error model to predict cyanobacterial biovolume from spring total phosphorus in Lake Mendota, Wisconsin

National Research Council Canada
Ottawa, Canada K1A 0R6

Conseil national de recherches Canada
Ottawa, Canada K1A 0R6

Canadian Journal of Fisheries and Aquatic Sciences

Volume 54, Number 2, February 1997

Journal canadien des sciences halieutiques et aquatiques

Volume 54, numéro 2, février 1997

	ARTICLES	ARTICLES
Bernard Sainte-Marie, Jean-Marie Sévigny, and Yves Gauthier	**239–248**	Laboratory behavior of adolescent and adult males of the snow crab (*Chionectes opilio*) (Brachyura: Majidae) mated noncompetitively and competitively with primiparous females
Martin J. Horgan and Edward L. Mills	**249–255**	Clearance rates and filtering activity of zebra mussel (*Dreissena polymorpha*): implications for freshwater lakes
Richard L. Leino and J. Howard McCormick	**256–263**	Reproductive characteristics of the ruffe, *Gymnocephalus cernuus*, in the St. Louis River estuary on western Lake Superior: a histological examination of the ovaries over one annual cycle
Jonathan D.S. Witt, Paul D.N. Hebert, and William B. Morton	**264–268**	*Echinogammarus ischnus*: another crustacean invader in the Laurentian Great Lakes basin
Linda A. Deegan, Bruce J. Peterson, Heidi Golden, Carole C. McIvor, and Michael C. Miller	**269–283**	Effects of fish density and river fertilization on algal standing stocks, invertebrate communities, and fish production in an arctic river
Murdoch K. McAllister and James N. Ianelli	**284–300**	Bayesian stock assessment using catch-age data and the sampling – importance resampling algorithm
Murdoch K. McAllister and Ellen K. Pikitch	**301–311**	A Bayesian approach to choosing a design for surveying fishery resources: application to the eastern Bering Sea trawl survey
Lee C. Bergstedt and Eric P. Bergersen	**312–319**	Health and movements of fish in response to sediment sluicing in the Wind River, Wyoming
Marley J. Waiser and Richard D. Robarts	**320–329**	Impacts of a herbicide and fertilizers on the microbial community of a saline prairie lake

Continued on inside back cover / Suite au verso

Canada

Printed in Canada by / Imprimé au Canada par
University of Toronto Press Inc.
Typesetting by / Typographie par
NRC Research Press / Presses scientifiques du CNRC

0706-652X(199702)54:2;1-C